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PCT/GB2005/000770

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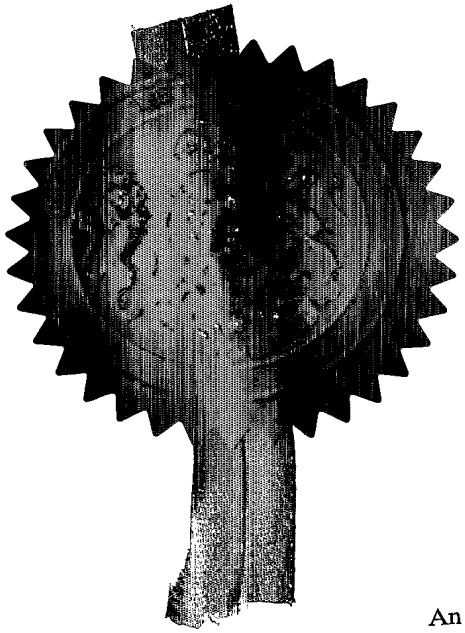

Signed William Morell

Dated 22 March 2005

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2. Patent application number
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3. Full name, address and postcode of the or of each applicant *(underline all surnames)*

SPIROGEN LIMITED
79 George Street
Ryde
ISLE OF WIGHT
PO33 2JF

08051872001

Patents ADP number *(if you know it)*

If the applicant is a corporate body, give the country/state of its incorporation

England

4. Title of the invention

PYRROLOBENZODIAZEPINES

5. Name of your agent *(if you have one)*

"Address for service" in the United Kingdom to which all correspondence should be sent *(including the postcode)*

MEWBURN ELLIS
York House
23 Kingsway
London WC2B 6HP

Patents ADP number *(if you know it)*

109006

6. Priority: Complete this section if you are declaring priority from one or more earlier patent applications, filed in the last 12 months.

Country	Priority application number *(if you know it)*	Date of filing *(day / month / year)*

7. Divisionals, etc: Complete this section only if this application is a divisional application or resulted from an entitlement dispute (see note f)

Number of earlier UK application	Date of filing *(day / month / year)*

8. Is a Patents Form 7/77 (Statement of inventorship and of right to grant of a patent) required in support of this request?

YES

Answer YES if:

a) any applicant named in part 3 is not an inventor, or
b) there is an inventor who is not named as an applicant, or
c) any named applicant is a corporate body.

Otherwise answer NO (See note d)

Patents Form 1/77

9. Accompanying documents: A patent application must include a description of the invention. Not counting duplicates, please enter the number of pages of each item accompanying this form:

Continuation sheets of this form	
Description	83
Claim(s)	
Abstract	SN
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10. If you are also filing any of the following, state how many against each item.

Priority documents	
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Statement of inventorship and right to grant of a patent (Patents Form 7/77)	
Request for a preliminary examination and search (Patents Form 9/77)	
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11. I/We request the grant of a patent on the basis of this application.

Signature(s)  Date 1 March 2004

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ROBERT WATSON
020 7240 4405

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e) Once you have filled in the form you must remember to sign and date it.

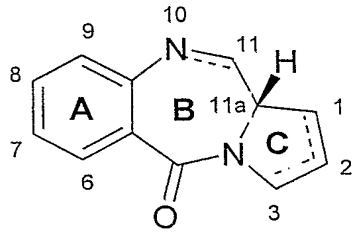
f) Part 7 should only be completed when a divisional application is being made under section 15(4), or when an application is being made under section 8(3), 12(6) or 37(4) following an entitlement dispute. By completing part 7 you are requesting that this application takes the same filing date as an earlier UK application. If you want the new application to have the same priority date(s) as the earlier UK application, you should also complete part 6 with the priority details.

PYRROLOBENZODIAZEPINES

The present invention relates to pyrrolobenzodiazepines (PBDs), and in particular pyrrolobenzodiazepines useful in the synthesis of dimeric compounds.

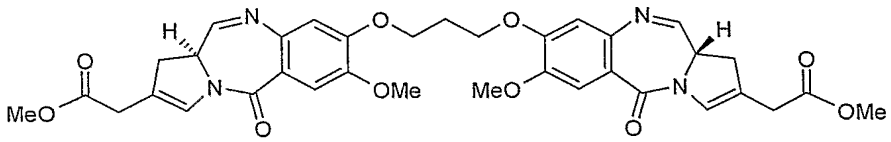
Background to the invention

Some pyrrolobenzodiazepines (PBDs) have the ability to recognise and bond to specific sequences of DNA; the preferred sequence is PuGPu. The first PBD antitumour antibiotic, anthramycin, was discovered in 1965 (Leimgruber, *et al., J. Am. Chem. Soc.*, **87**, 5793-5795 (1965); Leimgruber, *et al., J. Am. Chem. Soc.*, **87**, 5791-5793 (1965)). Since then, a number of naturally occurring PBDs have been reported, and over 10 synthetic routes have been developed to a variety of analogues (Thurston, *et al., Chem. Rev.* **1994**, 433-465 (1994)). Family members include abbeymycin (Hochlowski, *et al., J. Antibiotics*, **40**, 145-148 (1987)), chicamycin (Konishi, *et al., J. Antibiotics*, **37**, 200-206 (1984)), DC-81 (Japanese Patent 58-180 487; Thurston, *et al., Chem. Brit.*, **26**, 767-772 (1990); Bose, *et al., Tetrahedron*, **48**, 751-758 (1992)), mazethramycin (Kuminoto, *et al., J. Antibiotics*, **33**, 665-667 (1980)), neothramycins A and B (Takeuchi, *et al., J. Antibiotics*, **29**, 93-96 (1976)), porothramycin (Tsunakawa, *et al., J. Antibiotics*, **41**, 1366-1373 (1988)), prothracarcin (Shimizu, *et al, J. Antibiotics*, **29**, 2492-2503 (1982); Langley and Thurston, *J. Org. Chem.*, **52**, 91-97 (1987)), sibanomicin (DC-102)(Hara, *et al., J. Antibiotics*, **41**, 702-704 (1988); Itoh, *et al., J. Antibiotics*, **41**, 1281-1284 (1988)), sibiromycin (Leber, *et al., J. Am. Chem. Soc.*, **110**, 2992-2993 (1988)) and tomamycin (Arima, *et al., J. Antibiotics*, **25**, 437-444 (1972)). PBDs are of the general structure:



They differ in the number, type and position of substituents, in both their aromatic A rings and pyrrolo C rings, and in the degree of saturation of the C ring. In the B-ring there is either an imine (N=C), a carbinolamine(NH-CH(OH)), or a carbinolamine methyl ether (NH-CH(OMe)) at the N10-C11 position which is the electrophilic centre responsible for alkylating DNA. All of the known natural products have an (*S*)-configuration at the chiral C11a position which provides them with a right-handed twist when viewed from the C ring towards the A ring. This gives them the appropriate three-dimensional shape for isohelicity with the minor groove of B-form DNA, leading to a snug fit at the binding site (Kohn, In *Antibiotics III*. Springer-Verlag, New York, pp. 3-11 (1975); Hurley and Needham-VanDevanter, *Acc. Chem. Res.*, **19**, 230-237 (1986)). Their ability to form an adduct in the minor groove, enables them to interfere with DNA processing, hence their use as antitumour agents.

The present inventors have previously disclosed, in WO 00/12508, dimeric cytotoxic PBD compounds substituted at the C2 position, for example:



The synthesis of these compounds was achieved by formation of the dimeric backbone comprising the assembled A and C rings linked through the A ring by the diether linking chain. The N10 position was then protected with an Alloc group before a ring closure reaction to form the B ring and subsequent deprotection to give the product. The key stage in this synthesis is described as the

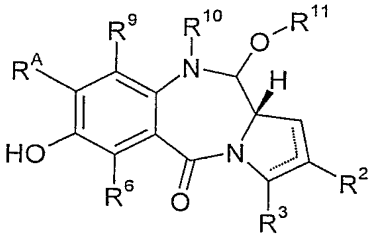
ring closure to form the B ring which occurs after the linking of the two A rings with the diether chain.

Using this route, to synthesise a number of dimers having the same monomer groups but different bridging groups require the synthesis of each compound from scratch, i.e. the synthesis route is not able to readily produce a diverse collection of PBD dimers, where the diversity is in the dimer bridge.

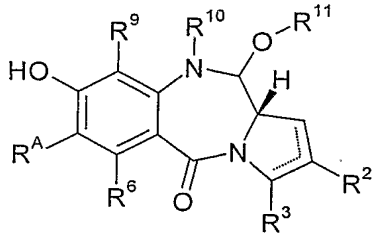
Disclosure of the invention

The present inventors have developed a key intermediate for the production of dimeric PBDs, which has a hydroxyl group at either the R^8 and/or R^7 position, a carbamate protecting group at the N10 position and a protected hydroxy group at the C11 position.

In a first aspect, the present invention comprises a compound with the formula **Ia** or **Ib**:



Ia



Ib

wherein:

the dotted lines indicate the optional presence of a double bond between C1 and C2 or C2 and C3;

R^2 and R^3 are independently selected from -H, =O, $=CH_2$, -CN, -R, OR, halo, =CH-R, $O-SO_2-R$, CO_2R and COR;

R^6 and R^9 are independently selected from H, R, OH, OR, SH, SR, NH_2, NHR, NRR', nitro, Me_3Sn and halo;

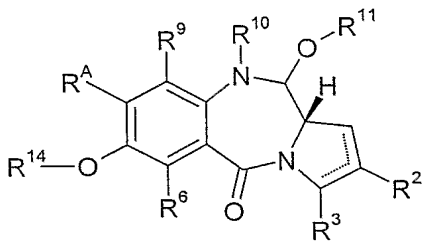
where R and R' are independently selected from optionally substituted C_{1-12} alkyl, C_{3-20} heterocyclyl and C_{5-20} aryl groups;

R^A is selected from H, R, OR, SH, SR, NH_2, NHR, NHRR', nitro, Me_3Sn and halo;

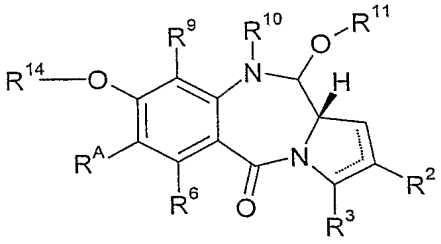
R^{10} is a carbamate-based nitrogen protecting group;

R^{11} is an oxygen protecting group.

In a second aspect, the present invention comprises a method of synthesising a compound of formula **Ia** or **Ib** as defined in the first aspect of the invention from a compound of formula **IIa** or **IIb** respectively:



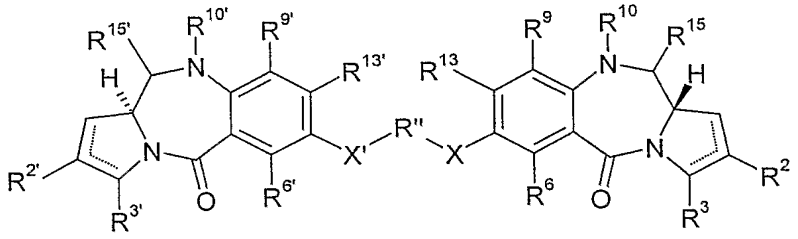
IIa



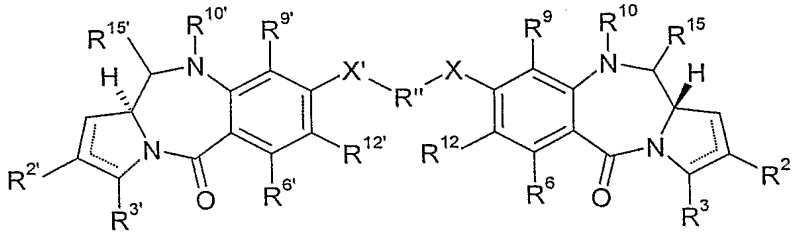
IIb

wherein R^A, R^2, R^3, R^6, R^9, R^{10} and R^{11} are as defined in the first aspect; R^{14} is an oxygen protecting group which is orthogonal to R^{11}.

In a third aspect, the present invention comprises a method of synthesising a compound of formula **IIIa** or **IIIb**:



IIIa



IIIb

or a solvate thereof, from a compound of formula **Ia** or **Ib** as defined in the first aspect, wherein R^6, R^7, R^8, R^9, R^{12} and R^{13} are as defined in the first aspect;

R^{10} is as defined in the first aspect and R^{15} is either O-R^{11}, wherein R^{11} is as defined in the first aspect, or OH, or R^{10} and R^{15} together form a double bond between N10 and C11; and

where R" is a C_{3-12} alkylene group, which chain may be interrupted by one or more heteroatoms, e.g. O, S, NH, and/or aromatic rings, e.g. benzene or pyridine, and each X is independently selected from O, S, or NH;

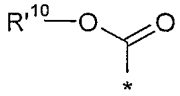
$R^{2'}$, $R^{3'}$, $R^{6'}$, $R^{9'}$, $R^{10'}$, $R^{12'}$ and $R^{15'}$ are all independently selected from the same lists as previously defined for R^2, R^3, R^6, R^9, R^{10}, R^{12} and R^{15} respectively.

Further aspects of the present invention relate to novel compounds of formula **IIIa** or **IIIb** (including solvates thereof when R^{10} and R^{15} form a double bond between N10 and C11, and pharmaceutical salts thereof), their use in methods of therapy (particularly in treating proliferative diseases), pharmaceutical compositions comprising these, and their use in the manufacture of a medicament for the treatment of a proliferative disease.

Definitions

Carbamate-based nitrogen protecting groups

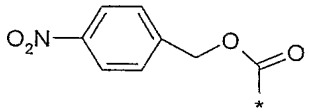
Carbamate-based nitrogen protecting groups are well known in the art, and have the following structure:



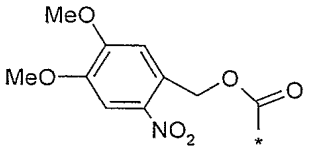
wherein R'^{10} is R as defined above. A large number of suitable groups are described on pages 503 to 549 of Greene, T.W. and Wuts, G.M., Protective Groups in Organic Synthesis, 3rd Edition, John Wiley & Sons, Inc., 1999, which is incorporated herein by reference.

Particularly preferred protecting groups include Alloc, Troc, Fmoc, CBz, Teoc, BOC, Doc, Hoc, TcBOC, 1-Adoc and 2-Adoc.

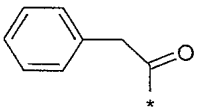
Also suitable for use in the present invention are nitrogen protecting group which can be removed *in vivo* (e.g. enzymatically, using light) as described in WO 00/12507, which is incorporated herein by reference. Examples of these protecting groups include:



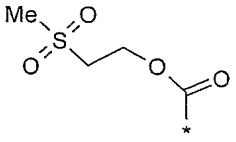
, which is nitroreductase labile (e.g. using ADEPT/GDEPT);



and



, which are photolabile; and



which is glutathione labile (e.g. using NPEPT).

Oxygen protecting groups

Oxygen protecting groups are well known in the art. A large number of suitable groups are described on pages 23 to 200 of Greene, T.W. and Wuts, G.M., Protective Groups in Organic Synthesis, 3rd Edition, John Wiley & Sons, Inc., 1999, which is incorporated herein by reference.

Classes of particular interest include silyl ethers, methyl ethers, alkyl ethers, benzyl ethers, esters, benzoates, carbonates, and sulfonates.

Preferred oxygen protecting groups include TBS, THP for the C11 oxygen atom, and benzyl ether for the C7 or C8 oxygen atom (where present).

As mentioned above the oxygen protecting group R^{14} should be orthogonal to the oxygen protecting group R^{11}. Protecting groups which are orthogonal to one another may each be removed using reagents or conditions which do not remove the other protecting group.

It may also be preferred that any protecting groups used during the synthesis and use of compounds of formula **I** are orthogonal to one another. However, it is often not necessary, but may be desirable, for the carbamate-based nitrogen protecting group and R^{11} to be orthogonal to one another, depending on whether the compound of formula **IIIa** or **IIIb** is to be used with the nitrogen protecting group in place.

Substituents

The phrase "optionally substituted" as used herein, pertains to a parent group which may be unsubstituted or which may be substituted.

Unless otherwise specified, the term "substituted" as used herein, pertains to a parent group which bears one or more substitutents. The term "substituent" is used herein in the conventional sense and refers to a chemical moiety which is covalently attached to, or if appropriate, fused to, a parent group. A wide variety of substituents are well known, and methods for their formation and introduction into a variety of parent groups are also well known.

Examples of substituents are described in more detail below.

C_{1-12} alkyl: The term "C_{1-12} alkyl" as used herein, pertains to a monovalent moiety obtained by removing a hydrogen atom from a carbon atom of a hydrocarbon compound having from 1 to 12 carbon atoms, which may be aliphatic or alicyclic, and which may be saturated or unsaturated (e.g. partially unsaturated, fully unsaturated). Thus, the term "alkyl" includes the sub-classes alkenyl, alkynyl, cycloalkyl, etc., discussed below.

Examples of saturated alkyl groups include, but are not limited to, methyl (C_1), ethyl (C_2), propyl (C_3), butyl (C_4), pentyl (C_5), hexyl (C_6) and heptyl (C_7).

Examples of saturated linear alkyl groups include, but are not limited to, methyl (C_1), ethyl (C_2), n-propyl (C_3), n-butyl (C_4), n-pentyl (amyl) (C_5), n-hexyl (C_6) and n-heptyl (C_7).

Examples of saturated branched alkyl groups include iso-propyl (C_3), iso-butyl (C_4), sec-butyl (C_4), tert-butyl (C_4), iso-pentyl (C_5), and neo-pentyl (C_5).

C_{2-12} Alkenyl: The term "C_{2-12} alkenyl" as used herein, pertains to an alkyl group having one or more carbon-carbon double bonds.

Examples of unsaturated alkenyl groups include, but are not limited to, ethenyl (vinyl, $-CH=CH_2$), 1-propenyl ($-CH=CH-CH_3$), 2-propenyl (allyl, $-CH-CH=CH_2$), isopropenyl (1-methylvinyl, $-C(CH_3)=CH_2$), butenyl (C_4), pentenyl (C_5), and hexenyl (C_6).

C_{2-12} alkynyl: The term "C_{2-12} alkynyl" as used herein, pertains to an alkyl group having one or more carbon-carbon triple bonds.

Examples of unsaturated alkynyl groups include, but are not limited to, ethynyl (ethinyl, $-C\equiv CH$) and 2-propynyl (propargyl, $-CH_2-C\equiv CH$).

C_{3-12} cycloalkyl: The term "C_{3-12} cycloalkyl" as used herein, pertains to an alkyl group which is also a cyclyl group; that is, a monovalent moiety obtained by removing a hydrogen atom from an alicyclic ring atom of a cyclic hydrocarbon (carbocyclic) compound, which moiety has from 3 to 7 carbon atoms, including from 3 to 7 ring atoms.

Examples of cycloalkyl groups include, but are not limited to, those derived from:

saturated monocyclic hydrocarbon compounds:

cyclopropane (C_3), cyclobutane (C_4), cyclopentane (C_5), cyclohexane (C_6), cycloheptane (C_7), methylcyclopropane (C_4), dimethylcyclopropane (C_5), methylcyclobutane (C_5),

dimethylcyclobutane (C_6), methylcyclopentane (C_6), dimethylcyclopentane (C_7) and methylcyclohexane (C_7);

unsaturated monocyclic hydrocarbon compounds: cyclopropene (C_3), cyclobutene (C_4), cyclopentene (C_5), cyclohexene (C_6), methylcyclopropene (C_4), dimethylcyclopropene (C_5), methylcyclobutene (C_5), dimethylcyclobutene (C_6), methylcyclopentene (C_6), dimethylcyclopentene (C_7) and methylcyclohexene (C_7); and

saturated polycyclic hydrocarbon compounds: norcarane (C_7), norpinane (C_7), norbornane (C_7).

C_{3-20} heterocyclyl: The term "C_{3-20} heterocyclyl" as used herein, pertains to a monovalent moiety obtained by removing a hydrogen atom from a ring atom of a heterocyclic compound, which moiety has from 3 to 20 ring atoms, of which from 1 to 10 are ring heteroatoms. Preferably, each ring has from 3 to 7 ring atoms, of which from 1 to 4 are ring heteroatoms.

In this context, the prefixes (e.g. C_{3-20}, C_{3-7}, C_{5-6}, etc.) denote the number of ring atoms, or range of number of ring atoms, whether carbon atoms or heteroatoms. For example, the term "C_{5-6}heterocyclyl", as used herein, pertains to a heterocyclyl group having 5 or 6 ring atoms.

Examples of monocyclic heterocyclyl groups include, but are not limited to, those derived from:

N_1: aziridine (C_3), azetidine (C_4), pyrrolidine (tetrahydropyrrole) (C_5), pyrroline (e.g., 3-pyrroline, 2,5-dihydropyrrole) (C_5), 2H-pyrrole or 3H-pyrrole (isopyrrole, isoazole) (C_5), piperidine (C_6), dihydropyridine (C_6), tetrahydropyridine (C_6), azepine (C_7);

O_1: oxirane (C_3), oxetane (C_4), oxolane (tetrahydrofuran) (C_5), oxole (dihydrofuran) (C_5), oxane (tetrahydropyran) (C_6), dihydropyran (C_6), pyran (C_6), oxepin (C_7);

S_1: thiirane (C_3), thietane (C_4), thiolane (tetrahydrothiophene) (C_5), thiane (tetrahydrothiopyran) (C_6), thiepane (C_7);

O_2: dioxolane (C_5), dioxane (C_6), and dioxepane (C_7);

O_3: trioxane (C_6);

N_2: imidazolidine (C_5), pyrazolidine (diazolidine) (C_5), imidazoline (C_5), pyrazoline (dihydropyrazole) (C_5), piperazine (C_6);

N_1O_1: tetrahydrooxazole (C_5), dihydrooxazole (C_5), tetrahydroisoxazole (C_5), dihydroisoxazole (C_5), morpholine (C_6), tetrahydrooxazine (C_6), dihydrooxazine (C_6), oxazine (C_6);

N_1S_1: thiazoline (C_5), thiazolidine (C_5), thiomorpholine (C_6);

N_2O_1: oxadiazine (C_6);

O_1S_1: oxathiole (C_5) and oxathiane (thioxane) (C_6); and,

$N_1O_1S_1$: oxathiazine (C_6).

Examples of substituted monocyclic heterocyclyl groups include those derived from saccharides, in cyclic form, for example, furanoses (C_5), such as arabinofuranose, lyxofuranose, ribofuranose, and xylofuranse, and pyranoses (C_6), such as allopyranose, altropyranose, glucopyranose, mannopyranose, gulopyranose, idopyranose, galactopyranose, and talopyranose.

C_{5-20} aryl: The term "C_{5-20} aryl", as used herein, pertains to a monovalent moiety obtained by removing a hydrogen atom from an aromatic ring atom of an aromatic compound, which moiety has from 3 to 20 ring atoms. Preferably, each ring has from 5 to 7 ring atoms.

In this context, the prefixes (e.g. C_{3-20}, C_{5-7}, C_{5-6}, etc.) denote the number of ring atoms, or range of number of ring atoms, whether carbon atoms or heteroatoms. For example, the term "C_{5-6} aryl" as used herein, pertains to an aryl group having 5 or 6 ring atoms.

The ring atoms may be all carbon atoms, as in "carboaryl groups". Examples of carboaryl groups include, but are not limited to, those derived from benzene (i.e. phenyl) (C_6), naphthalene (C_{10}), azulene (C_{10}), anthracene (C_{14}), phenanthrene (C_{14}), naphthacene (C_{18}), and pyrene (C_{16}).

Examples of aryl groups which comprise fused rings, at least one of which is an aromatic ring, include, but are not limited to, groups derived from indane (e.g. 2,3-dihydro-1H-indene) (C_9), indene (C_9), isoindene (C_9), tetraline (1,2,3,4-tetrahydronaphthalene (C_{10}), acenaphthene (C_{12}), fluorene (C_{13}), phenalene (C_{13}), acephenanthrene (C_{15}), and aceanthrene (C_{16}).

Alternatively, the ring atoms may include one or more heteroatoms, as in "heteroaryl groups". Examples of monocyclic heteroaryl groups include, but are not limited to, those derived from:
N_1: pyrrole (azole) (C_5), pyridine (azine) (C_6);
O_1: furan (oxole) (C_5);
S_1: thiophene (thiole) (C_5);
N_1O_1: oxazole (C_5), isoxazole (C_5), isoxazine (C_6);
N_2O_1: oxadiazole (furazan) (C_5);
N_3O_1: oxatriazole (C_5);
N_1S_1: thiazole (C_5), isothiazole (C_5);
N_2: imidazole (1,3-diazole) (C_5), pyrazole (1,2-diazole) (C_5), pyridazine (1,2-diazine) (C_6), pyrimidine (1,3-diazine) (C_6) (e.g., cytosine, thymine, uracil), pyrazine (1,4-diazine) (C_6);
N_3: triazole (C_5), triazine (C_6); and,
N_4: tetrazole (C_5).

Examples of heteroaryl which comprise fused rings, include, but are not limited to:

C_9 (with 2 fused rings) derived from benzofuran (O_1), isobenzofuran (O_1), indole (N_1), isoindole (N_1), indolizine (N_1), indoline (N_1), isoindoline (N_1), purine (N_4) (e.g., adenine, guanine), benzimidazole (N_2), indazole (N_2), benzoxazole (N_1O_1), benzisoxazole (N_1O_1), benzodioxole (O_2), benzofurazan (N_2O_1), benzotriazole (N_3), benzothiofuran (S_1), benzothiazole (N_1S_1), benzothiadiazole (N_2S);

C_{10} (with 2 fused rings) derived from chromene (O_1), isochromene (O_1), chroman (O_1), isochroman (O_1), benzodioxan (O_2), quinoline (N_1), isoquinoline (N_1), quinolizine (N_1), benzoxazine

(N_1O_1), benzodiazine (N_2), pyridopyridine (N_2), quinoxaline (N_2), quinazoline (N_2), cinnoline (N_2), phthalazine (N_2), naphthyridine (N_2), pteridine (N_4);

C_{11} (with 2 fused rings) derived from benzodiazepine (N_2);

C_{13} (with 3 fused rings) derived from carbazole (N_1), dibenzofuran (O_1), dibenzothiophene (S_1), carboline (N_2), perimidine (N_2), pyridoindole (N_2); and,

C_{14} (with 3 fused rings) derived from acridine (N_1), xanthene (O_1), thioxanthene (S_1), oxanthrene (O_2), phenoxathiin (O_1S_1), phenazine (N_2), phenoxazine (N_1O_1), phenothiazine (N_1S_1), thianthrene (S_2), phenanthridine (N_1), phenanthroline (N_2), phenazine (N_2).

The above groups, whether alone or part of another substituent, may themselves optionally be substituted with one or more groups selected from themselves and the additional substituents listed below.

Halo: -F, -Cl, -Br, and -I.

Hydroxy: -OH.

Ether: -OR, wherein R is an ether substituent, for example, a C_{1-7} alkyl group (also referred to as a C_{1-7} alkoxy group, discussed below), a C_{3-20} heterocyclyl group (also referred to as a C_{3-20} heterocyclyloxy group), or a C_{5-20} aryl group (also referred to as a C_{5-20} aryloxy group), preferably a C_{1-7}alkyl group.

Alkoxy: -OR, wherein R is an alkyl group, for example, a C_{1-7} alkyl group. Examples of C_{1-7} alkoxy groups include, but are not limited to, -OMe (methoxy), -OEt (ethoxy), -O(nPr) (n-propoxy), -O(iPr) (isopropoxy), -O(nBu) (n-butoxy), -O(sBu) (sec-butoxy), -O(iBu) (isobutoxy), and -O(tBu) (tert-butoxy).

Acetal: $-CH(OR^1)(OR^2)$, wherein R^1 and R^2 are independently acetal substituents, for example, a C_{1-7} alkyl group, a C_{3-20} heterocyclyl

group, or a C_{5-20} aryl group, preferably a C_{1-7} alkyl group, or, in the case of a "cyclic" acetal group, R^1 and R^2, taken together with the two oxygen atoms to which they are attached, and the carbon atoms to which they are attached, form a heterocyclic ring having from 4 to 8 ring atoms. Examples of acetal groups include, but are not limited to, $-CH(OMe)_2$, $-CH(OEt)_2$, and $-CH(OMe)(OEt)$.

Hemiacetal: $-CH(OH)(OR^1)$, wherein R^1 is a hemiacetal substituent, for example, a C_{1-7} alkyl group, a C_{3-20} heterocyclyl group, or a C_{5-20} aryl group, preferably a C_{1-7} alkyl group. Examples of hemiacetal groups include, but are not limited to, $-CH(OH)(OMe)$ and $-CH(OH)(OEt)$.

Ketal: $-CR(OR^1)(OR^2)$, where R^1 and R^2 are as defined for acetals, and R is a ketal substituent other than hydrogen, for example, a C_{1-7} alkyl group, a C_{3-20} heterocyclyl group, or a C_{5-20} aryl group, preferably a C_{1-7} alkyl group. Examples ketal groups include, but are not limited to, $-C(Me)(OMe)_2$, $-C(Me)(OEt)_2$, $-C(Me)(OMe)(OEt)$, $-C(Et)(OMe)_2$, $-C(Et)(OEt)_2$, and $-C(Et)(OMe)(OEt)$.

Hemiketal: $-CR(OH)(OR^1)$, where R^1 is as defined for hemiacetals, and R is a hemiketal substituent other than hydrogen, for example, a C_{1-7} alkyl group, a C_{3-20} heterocyclyl group, or a C_{5-20} aryl group, preferably a C_{1-7} alkyl group. Examples of hemiacetal groups include, but are not limited to, $-C(Me)(OH)(OMe)$, $-C(Et)(OH)(OMe)$, $-C(Me)(OH)(OEt)$, and $-C(Et)(OH)(OEt)$.

Oxo (keto, -one): =O.

Thione (thioketone): =S.

Imino (imine): =NR, wherein R is an imino substituent, for example, hydrogen, C_{1-7} alkyl group, a C_{3-20} heterocyclyl group, or a C_{5-20} aryl group, preferably hydrogen or a C_{1-7} alkyl group. Examples of ester groups include, but are not limited to, =NH, =NMe, =NEt, and =NPh.

Formyl (carbaldehyde, carboxaldehyde): -C(=O)H.

Acyl (keto): -C(=O)R, wherein R is an acyl substituent, for example, a C_{1-7} alkyl group (also referred to as C_{1-7} alkylacyl or C_{1-7} alkanoyl), a C_{3-20} heterocyclyl group (also referred to as C_{3-20} heterocyclylacyl), or a C_{5-20} aryl group (also referred to as C_{5-20} arylacyl), preferably a C_{1-7} alkyl group. Examples of acyl groups include, but are not limited to, -C(=O)CH_3 (acetyl), -C(=O)CH_2CH_3 (propionyl), -C(=O)$C(CH_3)_3$ (t-butyryl), and -C(=O)Ph (benzoyl, phenone).

Carboxy (carboxylic acid): -C(=O)OH.

Thiocarboxy (thiocarboxylic acid): -C(=S)SH.

Thiolocarboxy (thiolocarboxylic acid): -C(=O)SH.

Thionocarboxy (thionocarboxylic acid): -C(=S)OH.

Imidic acid: -C(=NH)OH.

Hydroxamic acid: -C(=NOH)OH.

Ester (carboxylate, carboxylic acid ester, oxycarbonyl): -C(=O)OR, wherein R is an ester substituent, for example, a C_{1-7} alkyl group, a C_{3-20} heterocyclyl group, or a C_{5-20} aryl group, preferably a C_{1-7} alkyl group. Examples of ester groups include, but are not limited to, -C(=O)OCH_3, -C(=O)OCH_2CH_3, -C(=O)$OC(CH_3)_3$, and -C(=O)OPh.

Acyloxy (reverse ester): -OC(=O)R, wherein R is an acyloxy substituent, for example, a C_{1-7} alkyl group, a C_{3-20} heterocyclyl group, or a C_{5-20} aryl group, preferably a C_{1-7} alkyl group. Examples of acyloxy groups include, but are not limited to, -OC(=O)CH_3

(acetoxy), $-OC(=O)CH_2CH_3$, $-OC(=O)C(CH_3)_3$, $-OC(=O)Ph$, and $-OC(=O)CH_2Ph$.

Oxycarboyloxy: $-OC(=O)OR$, wherein R is an ester substituent, for example, a C_{1-7} alkyl group, a C_{3-20} heterocyclyl group, or a C_{5-20} aryl group, preferably a C_{1-7} alkyl group. Examples of ester groups include, but are not limited to, $-OC(=O)OCH_3$, $-OC(=O)OCH_2CH_3$, $-OC(=O)OC(CH_3)_3$, and $-OC(=O)OPh$.

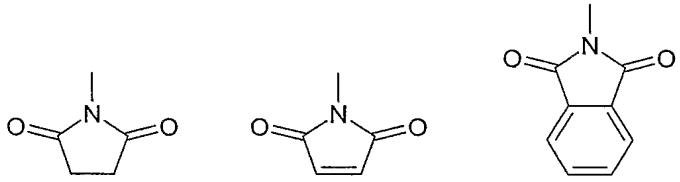
Amino: $-NR^1R^2$, wherein R^1 and R^2 are independently amino substituents, for example, hydrogen, a C_{1-7} alkyl group (also referred to as C_{1-7} alkylamino or di-C_{1-7} alkylamino), a C_{3-20} heterocyclyl group, or a C_{5-20} aryl group, preferably H or a C_{1-7} alkyl group, or, in the case of a "cyclic" amino group, R^1 and R^2, taken together with the nitrogen atom to which they are attached, form a heterocyclic ring having from 4 to 8 ring atoms. Amino groups may be primary ($-NH_2$), secondary ($-NHR^1$), or tertiary ($-NHR^1R^2$), and in cationic form, may be quaternary ($-^+NR^1R^2R^3$). Examples of amino groups include, but are not limited to, $-NH_2$, $-NHCH_3$, $-NHC(CH_3)_2$, $-N(CH_3)_2$, $-N(CH_2CH_3)_2$, and -NHPh. Examples of cyclic amino groups include, but are not limited to, aziridino, azetidino, pyrrolidino, piperidino, piperazino, morpholino, and thiomorpholino.

Amido (carbamoyl, carbamyl, aminocarbonyl, carboxamide): $-C(=O)NR^1R^2$, wherein R^1 and R^2 are independently amino substituents, as defined for amino groups. Examples of amido groups include, but are not limited to, $-C(=O)NH_2$, $-C(=O)NHCH_3$, $-C(=O)N(CH_3)_2$, $-C(=O)NHCH_2CH_3$, and $-C(=O)N(CH_2CH_3)_2$, as well as amido groups in which R^1 and R^2, together with the nitrogen atom to which they are attached, form a heterocyclic structure as in, for example, piperidinocarbonyl, morpholinocarbonyl, thiomorpholinocarbonyl, and piperazinocarbonyl.

Thioamido (thiocarbamyl): $-C(=S)NR^1R^2$, wherein R^1 and R^2 are independently amino substituents, as defined for amino groups.

Examples of amido groups include, but are not limited to, $-C(=S)NH_2$, $-C(=S)NHCH_3$, $-C(=S)N(CH_3)_2$, and $-C(=S)NHCH_2CH_3$.

Acylamido (acylamino): $-NR^1C(=O)R^2$, wherein R^1 is an amide substituent, for example, hydrogen, a C_{1-7} alkyl group, a C_{3-20} heterocyclyl group, or a C_{5-20} aryl group, preferably hydrogen or a C_{1-7} alkyl group, and R^2 is an acyl substituent, for example, a C_{1-7} alkyl group, a C_{3-20} heterocyclyl group, or a C_{5-20}aryl group, preferably hydrogen or a C_{1-7} alkyl group. Examples of acylamide groups include, but are not limited to, $-NHC(=O)CH_3$, $-NHC(=O)CH_2CH_3$, and $-NHC(=O)Ph$. R^1 and R^2 may together form a cyclic structure, as in, for example, succinimidyl, maleimidyl, and phthalimidyl:

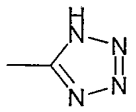


Aminocarbonyloxy: $-OC(=O)NR^1R^2$, wherein R^1 and R^2 are independently amino substituents, as defined for amino groups. Examples of aminocarbonyloxy groups include, but are not limited to, $-OC(=O)NH_2$, $-OC(=O)NHMe$, $-OC(=O)NMe_2$, and $-OC(=O)NEt_2$.

Ureido: $-N(R^1)CONR^2R^3$ wherein R^2 and R^3 are independently amino substituents, as defined for amino groups, and R^1 is a ureido substituent, for example, hydrogen, a C_{1-7} alkyl group, a C_{3-20} heterocyclyl group, or a C_{5-20} aryl group, preferably hydrogen or a C_{1-7} alkyl group. Examples of ureido groups include, but are not limited to, $-NHCONH_2$, $-NHCONHMe$, $-NHCONHEt$, $-NHCONMe_2$, $-NHCONEt_2$, $-NMeCONH_2$, $-NMeCONHMe$, $-NMeCONHEt$, $-NMeCONMe_2$, and $-NMeCONEt_2$.

Guanidino: $-NH-C(=NH)NH_2$.

Tetrazolyl: a five membered aromatic ring having four nitrogen atoms and one carbon atom,



Imino: =NR, wherein R is an imino substituent, for example, for example, hydrogen, a C_{1-7} alkyl group, a C_{3-20} heterocyclyl group, or a C_{5-20} aryl group, preferably H or a C_{1-7}alkyl group. Examples of imino groups include, but are not limited to, =NH, =NMe, and =NEt.

Amidine (amidino): $-C(=NR)NR_2$, wherein each R is an amidine substituent, for example, hydrogen, a C_{1-7} alkyl group, a C_{3-20} heterocyclyl group, or a C_{5-20} aryl group, preferably H or a C_{1-7} alkyl group. Examples of amidine groups include, but are not limited to, $-C(=NH)NH_2$, $-C(=NH)NMe_2$, and $-C(=NMe)NMe_2$.

Nitro: $-NO_2$.

Nitroso: -NO.

Azido: $-N_3$.

Cyano (nitrile, carbonitrile): -CN.

Isocyano: -NC.

Cyanato: -OCN.

Isocyanato: -NCO.

Thiocyano (thiocyanato): -SCN.

Isothiocyano (isothiocyanato): -NCS.

Sulfhydryl (thiol, mercapto): -SH.

Thioether (sulfide): -SR, wherein R is a thioether substituent, for example, a C_{1-7} alkyl group (also referred to as a C_{1-7}alkylthio group), a C_{3-20} heterocyclyl group, or a C_{5-20} aryl group, preferably a C_{1-7} alkyl group. Examples of C_{1-7} alkylthio groups include, but are not limited to, $-SCH_3$ and $-SCH_2CH_3$.

Disulfide: -SS-R, wherein R is a disulfide substituent, for example, a C_{1-7} alkyl group, a C_{3-20} heterocyclyl group, or a C_{5-20} aryl group, preferably a C_{1-7} alkyl group (also referred to herein as C_{1-7} alkyl disulfide). Examples of C_{1-7} alkyl disulfide groups include, but are not limited to, $-SSCH_3$ and $-SSCH_2CH_3$.

Sulfine (sulfinyl, sulfoxide): $-S(=O)R$, wherein R is a sulfine substituent, for example, a C_{1-7} alkyl group, a C_{3-20} heterocyclyl group, or a C_{5-20} aryl group, preferably a C_{1-7} alkyl group. Examples of sulfine groups include, but are not limited to, $-S(=O)CH_3$ and $-S(=O)CH_2CH_3$.

Sulfone (sulfonyl): $-S(=O)_2R$, wherein R is a sulfone substituent, for example, a C_{1-7} alkyl group, a C_{3-20} heterocyclyl group, or a C_{5-20} aryl group, preferably a C_{1-7} alkyl group, including, for example, a fluorinated or perfluorinated C_{1-7} alkyl group. Examples of sulfone groups include, but are not limited to, $-S(=O)_2CH_3$ (methanesulfonyl, mesyl), $-S(=O)_2CF_3$ (triflyl), $-S(=O)_2CH_2CH_3$ (esyl), $-S(=O)_2C_4F_9$ (nonaflyl), $-S(=O)_2CH_2CF_3$ (tresyl), $-S(=O)_2CH_2CH_2NH_2$ (tauryl), $-S(=O)_2Ph$ (phenylsulfonyl, besyl), 4-methylphenylsulfonyl (tosyl), 4-chlorophenylsulfonyl (closyl), 4-bromophenylsulfonyl (brosyl), 4-nitrophenyl (nosyl), 2-naphthalenesulfonate (napsyl), and 5-dimethylamino-naphthalen-1-ylsulfonate (dansyl).

Sulfinic acid (sulfino): $-S(=O)OH$, $-SO_2H$.

Sulfonic acid (sulfo): $-S(=O)_2OH$, $-SO_3H$.

Sulfinate (sulfinic acid ester): -S(=O)OR; wherein R is a sulfinate substituent, for example, a C_{1-7} alkyl group, a C_{3-20} heterocyclyl group, or a C_{5-20} aryl group, preferably a C_{1-7} alkyl group. Examples of sulfinate groups include, but are not limited to, $-S(=O)OCH_3$ (methoxysulfinyl; methyl sulfinate) and $-S(=O)OCH_2CH_3$ (ethoxysulfinyl; ethyl sulfinate).

Sulfonate (sulfonic acid ester): $-S(=O)_2OR$, wherein R is a sulfonate substituent, for example, a C_{1-7} alkyl group, a C_{3-20} heterocyclyl group, or a C_{5-20} aryl group, preferably a C_{1-7} alkyl group. Examples of sulfonate groups include, but are not limited to, $-S(=O)_2OCH_3$ (methoxysulfonyl; methyl sulfonate) and $-S(=O)_2OCH_2CH_3$ (ethoxysulfonyl; ethyl sulfonate).

Sulfinyloxy: $-OS(=O)R$, wherein R is a sulfinyloxy substituent, for example, a C_{1-7} alkyl group, a C_{3-20} heterocyclyl group, or a C_{5-20} aryl group, preferably a C_{1-7} alkyl group. Examples of sulfinyloxy groups include, but are not limited to, $-OS(=O)CH_3$ and $-OS(=O)CH_2CH_3$.

Sulfonyloxy: $-OS(=O)_2R$, wherein R is a sulfonyloxy substituent, for example, a C_{1-7} alkyl group, a C_{3-20} heterocyclyl group, or a C_{5-20} aryl group, preferably a C_{1-7} alkyl group. Examples of sulfonyloxy groups include, but are not limited to, $-OS(=O)_2CH_3$ (mesylate) and $-OS(=O)_2CH_2CH_3$ (esylate).

Sulfate: $-OS(=O)_2OR$; wherein R is a sulfate substituent, for example, a C_{1-7} alkyl group, a C_{3-20} heterocyclyl group, or a C_{5-20} aryl group, preferably a C_{1-7} alkyl group. Examples of sulfate groups include, but are not limited to, $-OS(=O)_2OCH_3$ and $-SO(=O)_2OCH_2CH_3$.

Sulfamyl (sulfamoyl; sulfinic acid amide; sulfinamide): $-S(=O)NR^1R^2$, wherein R^1 and R^2 are independently amino substituents, as defined for amino groups. Examples of sulfamyl groups include,

but are not limited to, $-S(=O)NH_2$, $-S(=O)NH(CH_3)$, $-S(=O)N(CH_3)_2$, $-S(=O)NH(CH_2CH_3)$, $-S(=O)N(CH_2CH_3)_2$, and $-S(=O)NHPh$.

Sulfonamido (sulfinamoyl; sulfonic acid amide; sulfonamide): $-S(=O)_2NR^1R^2$, wherein R^1 and R^2 are independently amino substituents, as defined for amino groups. Examples of sulfonamido groups include, but are not limited to, $-S(=O)_2NH_2$, $-S(=O)_2NH(CH_3)$, $-S(=O)_2N(CH_3)_2$, $-S(=O)_2NH(CH_2CH_3)$, $-S(=O)_2N(CH_2CH_3)_2$, and $-S(=O)_2NHPh$.

Sulfamino: $-NR^1S(=O)_2OH$, wherein R^1 is an amino substituent, as defined for amino groups. Examples of sulfamino groups include, but are not limited to, $-NHS(=O)_2OH$ and $-N(CH_3)S(=O)_2OH$.

Sulfonamino: $-NR^1S(=O)_2R$, wherein R^1 is an amino substituent, as defined for amino groups, and R is a sulfonamino substituent, for example, a C_{1-7} alkyl group, a C_{3-20} heterocyclyl group, or a C_{5-20} aryl group, preferably a C_{1-7} alkyl group. Examples of sulfonamino groups include, but are not limited to, $-NHS(=O)_2CH_3$ and $-N(CH_3)S(=O)_2C_6H_5$.

Sulfinamino: $-NR^1S(=O)R$, wherein R^1 is an amino substituent, as defined for amino groups, and R is a sulfinamino substituent, for example, a C_{1-7} alkyl group, a C_{3-20} heterocyclyl group, or a C_{5-20} aryl group, preferably a C_{1-7} alkyl group. Examples of sulfinamino groups include, but are not limited to, $-NHS(=O)CH_3$ and $-N(CH_3)S(=O)C_6H_5$.

Phosphino (phosphine): $-PR_2$, wherein R is a phosphino substituent, for example, -H, a C_{1-7} alkyl group, a C_{3-20} heterocyclyl group, or a C_{5-20} aryl group, preferably -H, a C_{1-7} alkyl group, or a C_{5-20} aryl group. Examples of phosphino groups include, but are not limited to, $-PH_2$, $-P(CH_3)_2$, $-P(CH_2CH_3)_2$, $-P(t\text{-}Bu)_2$, and $-P(Ph)_2$.

Phospho: $-P(=O)_2$.

Phosphinyl (phosphine oxide): $-P(=O)R_2$, wherein R is a phosphinyl substituent, for example, a C_{1-7} alkyl group, a C_{3-20} heterocyclyl group, or a C_{5-20} aryl group, preferably a C_{1-7} alkyl group or a C_{5-20} aryl group. Examples of phosphinyl groups include, but are not limited to, $-P(=O)(CH_3)_2$, $-P(=O)(CH_2CH_3)_2$, $-P(=O)(t\text{-}Bu)_2$, and $-P(=O)(Ph)_2$.

Phosphonic acid (phosphono): $-P(=O)(OH)_2$.

Phosphonate (phosphono ester): $-P(=O)(OR)_2$, where R is a phosphonate substituent, for example, -H, a C_{1-7} alkyl group, a C_{3-20} heterocyclyl group, or a C_{5-20} aryl group, preferably -H, a C_{1-7} alkyl group, or a C_{5-20} aryl group. Examples of phosphonate groups include, but are not limited to, $-P(=O)(OCH_3)_2$, $-P(=O)(OCH_2CH_3)_2$, $-P(=O)(O\text{-}t\text{-}Bu)_2$, and $-P(=O)(OPh)_2$.

Phosphoric acid (phosphonooxy): $-OP(=O)(OH)_2$.

Phosphate (phosphonooxy ester): $-OP(=O)(OR)_2$, where R is a phosphate substituent, for example, -H, a C_{1-7} alkyl group, a C_{3-20} heterocyclyl group, or a C_{5-20} aryl group, preferably -H, a C_{1-7} alkyl group, or a C_{5-20} aryl group. Examples of phosphate groups include, but are not limited to, $-OP(=O)(OCH_3)_2$, $-OP(=O)(OCH_2CH_3)_2$, $-OP(=O)(O\text{-}t\text{-}Bu)_2$, and $-OP(=O)(OPh)_2$.

Phosphorous acid: $-OP(OH)_2$.

Phosphite: $-OP(OR)_2$, where R is a phosphite substituent, for example, -H, a C_{1-7} alkyl group, a C_{3-20} heterocyclyl group, or a C_{5-20} aryl group, preferably -H, a C_{1-7} alkyl group, or a C_{5-20} aryl group. Examples of phosphite groups include, but are not limited to, $-OP(OCH_3)_2$, $-OP(OCH_2CH_3)_2$, $-OP(O\text{-}t\text{-}Bu)_2$, and $-OP(OPh)_2$.

Phosphoramidite: $-OP(OR^1)-NR^2_2$, where R^1 and R^2 are phosphoramidite substituents, for example, -H, a (optionally substituted) C_{1-7} alkyl group, a C_{3-20} heterocyclyl group, or a C_{5-20} aryl group, preferably -

H, a C_{1-7} alkyl group, or a C_{5-20} aryl group. Examples of phosphoramidite groups include, but are not limited to, $-OP(OCH_2CH_3)-N(CH_3)_2$, $-OP(OCH_2CH_3)-N(i-Pr)_2$, and $-OP(OCH_2CH_2CN)-N(i-Pr)_2$.

Phosphoramidate: $-OP(=O)(OR^1)-NR^2_2$, where R^1 and R^2 are phosphoramidate substituents, for example, -H, a (optionally substituted) C_{1-7} alkyl group, a C_{3-20} heterocyclyl group, or a C_{5-20} aryl group, preferably -H, a C_{1-7} alkyl group, or a C_{5-20} aryl group. Examples of phosphoramidate groups include, but are not limited to, $-OP(=O)(OCH_2CH_3)-N(CH_3)_2$, $-OP(=O)(OCH_2CH_3)-N(i-Pr)_2$, and $-OP(=O)(OCH_2CH_2CN)-N(i-Pr)_2$.

Alkylene

C_{3-12} alkylene: The term "C_{3-12} alkylene", as used herein, pertains to a bidentate moiety obtained by removing two hydrogen atoms, either both from the same carbon atom, or one from each of two different carbon atoms, of a hydrocarbon compound having from 3 to 12 carbon atoms (unless otherwise specified), which may be aliphatic or alicyclic, and which may be saturated, partially unsaturated, or fully unsaturated. Thus, the term "alkylene" includes the sub-classes alkenylene, alkynylene, cycloalkylene, etc., discussed below.

Examples of linear saturated C_{3-12} alkylene groups include, but are not limited to, $-(CH_2)_n-$ where n is an integer from 3 to 12, for example, $-CH_2CH_2CH_2-$ (propylene), $-CH_2CH_2CH_2CH_2-$ (butylene), $-CH_2CH_2CH_2CH_2CH_2-$ (pentylene) and $-CH_2CH_2CH_2CH_2CH_2CH_2CH_2-$ (heptylene).

Examples of branched saturated C_{3-12} alkylene groups include, but are not limited to, $-CH(CH_3)CH_2-$, $-CH(CH_3)CH_2CH_2-$, $-CH(CH_3)CH_2CH_2CH_2-$, $-CH_2CH(CH_3)CH_2-$, $-CH_2CH(CH_3)CH_2CH_2-$, $-CH(CH_2CH_3)-$, $-CH(CH_2CH_3)CH_2-$, and $-CH_2CH(CH_2CH_3)CH_2-$.

Examples of linear partially unsaturated C_{3-12} alkylene groups (C_{3-12} alkenylene, and alkynylene groups) include, but are not limited

to, -CH=CH-CH_2-, -CH_2-CH=CH_2-, -CH=CH-CH_2-CH_2-, -CH=CH-CH_2-CH_2-CH_2-, -CH=CH-CH=CH-, -CH=CH-CH=CH-CH_2-, -CH=CH-CH=CH-CH_2-CH_2-, -CH=CH-CH_2-CH=CH-, -CH=CH-CH_2-CH_2-CH=CH-, and -CH_2-C≡C-CH_2-.

Examples of branched partially unsaturated C_{3-12} alkylene groups (C_{3-12} alkenylene and alkynylene groups) include, but are not limited to, -C(CH_3)=CH-, -C(CH_3)=CH-CH_2-, -CH=CH-CH(CH_3)- and -C≡C-CH(CH_3)-.

Examples of alicyclic saturated C_{3-12} alkylene groups (C_{3-12} cycloalkylenes) include, but are not limited to, cyclopentylene (e.g. cyclopent-1,3-ylene), and cyclohexylene (e.g. cyclohex-1,4-ylene).

Examples of alicyclic partially unsaturated C_{3-12} alkylene groups (C_{3-12} cycloalkylenes) include, but are not limited to, cyclopentenylene (e.g. 4-cyclopenten-1,3-ylene), cyclohexenylene (e.g. 2-cyclohexen-1,4-ylene; 3-cyclohexen-1,2-ylene; 2,5-cyclohexadien-1,4-ylene).

Proliferative Diseases

One of ordinary skill in the art is readily able to determine whether or not a candidate compound treats a proliferative condition for any particular cell type. For example, assays which may conveniently be used to assess the activity offered by a particular compound are described in the examples below.

The term "proliferative disease" pertains to an unwanted or uncontrolled cellular proliferation of excessive or abnormal cells which is undesired, such as, neoplastic or hyperplastic growth, whether *in vitro* or *in vivo*.

Examples of proliferative conditions include, but are not limited to, benign, pre-malignant, and malignant cellular proliferation, including but not limited to, neoplasms and tumours (e.g. histocytoma, glioma, astrocyoma, osteoma), cancers (e.g. lung cancer, small cell lung cancer, gastrointestinal cancer, bowel cancer, colon cancer, breast carinoma, ovarian carcinoma, prostate

cancer, testicular cancer, liver cancer, kidney cancer, bladder cancer, pancreas cancer, brain cancer, sarcoma, osteosarcoma, Kaposi's sarcoma, melanoma), leukemias, psoriasis, bone diseases, fibroproliferative disorders (e.g. of connective tissues), and atherosclerosis.

Any type of cell may be treated, including but not limited to, lung, gastrointestinal (including, e.g. bowel, colon), breast (mammary), ovarian, prostate, liver (hepatic), kidney (renal), bladder, pancreas, brain, and skin.

Methods of Treatment

As described above, the present invention provide the use of a compound of formula **IIIa** or **IIIb** in a method of therapy. Also provided is a method of treatment, comprising administering to a subject in need of treatment a therapeutically-effective amount of a compound of formula **IIIa** or **IIIb**, preferably in the form of a pharmaceutical composition, which is the third aspect of the present invention. The term "therapeutically effective amount" is an amount sufficient to show benefit to a patient. Such benefit may be at least amelioration of at least one symptom. The actual amount administered, and rate and time-course of administration, will depend on the nature and severity of what is being treated. Prescription of treatment, e.g. decisions on dosage, is within the responsibility of general practitioners and other medical doctors.

A compound may be administered alone or in combination with other treatments, either simultaneously or sequentially dependent upon the condition to be treated. Examples of treatments and therapies include, but are not limited to, chemotherapy (the administration of active agents, including, e.g. drugs; surgery; and radiation therapy. If the compound of formula **IIIa** or **IIIb** bears a carbamate-based nitrogen protecting group which may be removed *in vivo*, then the methods of treatment described in WO 00/12507 (ADEPT, GDEPT and PDT) may be used.

Pharmaceutical compositions according to the present invention, and for use in accordance with the present invention, may comprise, in addition to the active ingredient, i.e. a compound of formula **IIIa** or **IIIb**, a pharmaceutically acceptable excipient, carrier, buffer, stabiliser or other materials well known to those skilled in the art. Such materials should be non-toxic and should not interfere with the efficacy of the active ingredient. The precise nature of the carrier or other material will depend on the route of administration, which may be oral, or by injection, e.g. cutaneous, subcutaneous, or intravenous.

Pharmaceutical compositions for oral administration may be in tablet, capsule, powder or liquid form. A tablet may comprise a solid carrier or an adjuvant. Liquid pharmaceutical compositions generally comprise a liquid carrier such as water, petroleum, animal or vegetable oils, mineral oil or synthetic oil. Physiological saline solution, dextrose or other saccharide solution or glycols such as ethylene glycol, propylene glycol or polyethylene glycol may be included. A capsule may comprise a solid carrier such a gelatin.

For intravenous, cutaneous or subcutaneous injection, or injection at the site of affliction, the active ingredient will be in the form of a parenterally acceptable aqueous solution which is pyrogen-free and has suitable pH, isotonicity and stability. Those of relevant skill in the art are well able to prepare suitable solutions using, for example, isotonic vehicles such as Sodium Chloride Injection, Ringer's Injection, Lactated Ringer's Injection. Preservatives, stabilisers, buffers, antioxidants and/or other additives may be included, as required.

Includes Other Forms

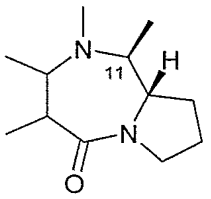
Unless otherwise specified, included in the above are the well known ionic, salt, solvate, and protected forms of these substituents. For example, a reference to carboxylic acid (-COOH) also includes the anionic (carboxylate) form ($-COO^-$), a salt or

solvate thereof, as well as conventional protected forms. Similarly, a reference to an amino group includes the protonated form ($-N^+HR^1R^2$), a salt or solvate of the amino group, for example, a hydrochloride salt, as well as conventional protected forms of an amino group. Similarly, a reference to a hydroxyl group also includes the anionic form ($-O^-$), a salt or solvate thereof, as well as conventional protected forms.

Isomers, Salts and Solvates

Certain compounds may exist in one or more particular geometric, optical, enantiomeric, diasteriomeric, epimeric, atropic, stereoisomeric, tautomeric, conformational, or anomeric forms, including but not limited to, cis- and trans-forms; E- and Z-forms; c-, t-, and r- forms; endo- and exo-forms; R-, S-, and meso-forms; D- and L-forms; d- and l-forms; (+) and (-) forms; keto-, enol-, and enolate-forms; syn- and anti-forms; synclinal- and anticlinal-forms; α- and β-forms; axial and equatorial forms; boat-, chair-, twist-, envelope-, and halfchair-forms; and combinations thereof, hereinafter collectively referred to as "isomers" (or "isomeric forms").

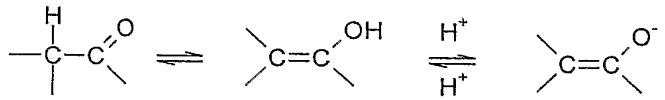
Preferably compounds of the present invention have the following stereochemistry at the C11 position:



Note that, except as discussed below for tautomeric forms, specifically excluded from the term "isomers", as used herein, are structural (or constitutional) isomers (i.e. isomers which differ in the connections between atoms rather than merely by the position of atoms in space). For example, a reference to a methoxy group, $-OCH_3$, is not to be construed as a reference to its structural isomer, a hydroxymethyl group, $-CH_2OH$. Similarly, a

reference to ortho-chlorophenyl is not to be construed as a reference to its structural isomer, meta-chlorophenyl. However, a reference to a class of structures may well include structurally isomeric forms falling within that class (e.g. C_{1-7} alkyl includes n-propyl and iso-propyl; butyl includes n-, iso-, sec-, and tert-butyl; methoxyphenyl includes ortho-, meta-, and para-methoxyphenyl).

The above exclusion does not pertain to tautomeric forms, for example, keto-, enol-, and enolate-forms, as in, for example, the following tautomeric pairs: keto/enol (illustrated below), imine/enamine, amide/imino alcohol, amidine/amidine, nitroso/oxime, thioketone/enethiol, N-nitroso/hyroxyazo, and nitro/aci-nitro.



keto enol enolate

Note that specifically included in the term "isomer" are compounds with one or more isotopic substitutions. For example, H may be in any isotopic form, including ^{1}H, ^{2}H (D), and ^{3}H (T); C may be in any isotopic form, including ^{12}C, ^{13}C, and ^{14}C; O may be in any isotopic form, including ^{16}O and ^{18}O; and the like.

Unless otherwise specified, a reference to a particular compound includes all such isomeric forms, including (wholly or partially) racemic and other mixtures thereof. Methods for the preparation (e.g. asymmetric synthesis) and separation (e.g. fractional crystallisation and chromatographic means) of such isomeric forms are either known in the art or are readily obtained by adapting the methods taught herein, or known methods, in a known manner.

Unless otherwise specified, a reference to a particular compound also includes ionic, salt, solvate, and protected forms of thereof, for example, as discussed below.

It may be convenient or desirable to prepare, purify, and/or handle a corresponding salt of the active compound, for example, a pharmaceutically-acceptable salt. Examples of pharmaceutically acceptable salts are discussed in Berge, *et al.*, *J. Pharm. Sci.*, **66**, 1-19 (1977).

For example, if the compound is anionic, or has a functional group which may be anionic (e.g. -COOH may be $-COO^-$), then a salt may be formed with a suitable cation. Examples of suitable inorganic cations include, but are not limited to, alkali metal ions such as Na^+ and K^+, alkaline earth cations such as Ca^{2+} and Mg^{2+}, and other cations such as Al^{+3}. Examples of suitable organic cations include, but are not limited to, ammonium ion (i.e. NH_4^+) and substituted ammonium ions (e.g. NH_3R^+, $NH_2R_2^+$, NHR_3^+, NR_4^+). Examples of some suitable substituted ammonium ions are those derived from: ethylamine, diethylamine, dicyclohexylamine, triethylamine, butylamine, ethylenediamine, ethanolamine, diethanolamine, piperazine, benzylamine, phenylbenzylamine, choline, meglumine, and tromethamine, as well as amino acids, such as lysine and arginine. An example of a common quaternary ammonium ion is $N(CH_3)_4^+$.

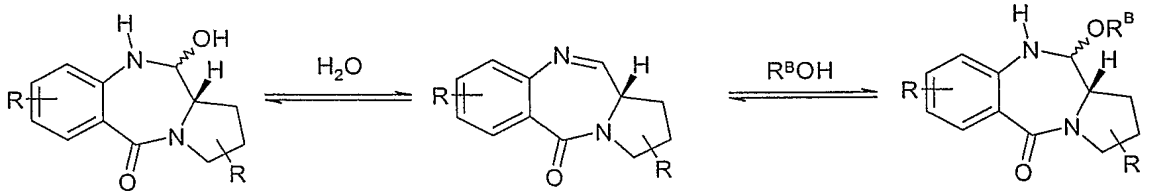
If the compound is cationic, or has a functional group which may be cationic (e.g. $-NH_2$ may be $-NH_3^+$), then a salt may be formed with a suitable anion. Examples of suitable inorganic anions include, but are not limited to, those derived from the following inorganic acids: hydrochloric, hydrobromic, hydroiodic, sulfuric, sulfurous, nitric, nitrous, phosphoric, and phosphorous.

Examples of suitable organic anions include, but are not limited to, those derived from the following organic acids: 2-acetyoxybenzoic, acetic, ascorbic, aspartic, benzoic, camphorsulfonic, cinnamic, citric, edetic, ethanedisulfonic, ethanesulfonic, fumaric, glucheptonic, gluconic, glutamic, glycolic, hydroxymaleic, hydroxynaphthalene carboxylic,

isethionic, lactic, lactobionic, lauric, maleic, malic, methanesulfonic, mucic, oleic, oxalic, palmitic, pamoic, pantothenic, phenylacetic, phenylsulfonic, propionic, pyruvic, salicylic, stearic, succinic, sulfanilic, tartaric, toluenesulfonic, and valeric. Examples of suitable polymeric organic anions include, but are not limited to, those derived from the following polymeric acids: tannic acid, carboxymethyl cellulose.

It may be convenient or desirable to prepare, purify, and/or handle a corresponding solvate of the active compound. The term "solvate" is used herein in the conventional sense to refer to a complex of solute (e.g. active compound, salt of active compound) and solvent. If the solvent is water, the solvate may be conveniently referred to as a hydrate, for example, a mono-hydrate, a di-hydrate, a tri-hydrate, etc.

Solvates of particular relevance to the present invention are those where the solvent adds across the imine bond of the PBD moiety, which is illustrated below where the solvent is water or an alcohol (R^BOH, where R^B is an ether substituent as described above):



These forms can be called the carbinolamine and carbinolamine ether forms of the PBD. The balance of these equilibria depend on the conditions in which the compounds are found, as well as the nature of the moiety itself.

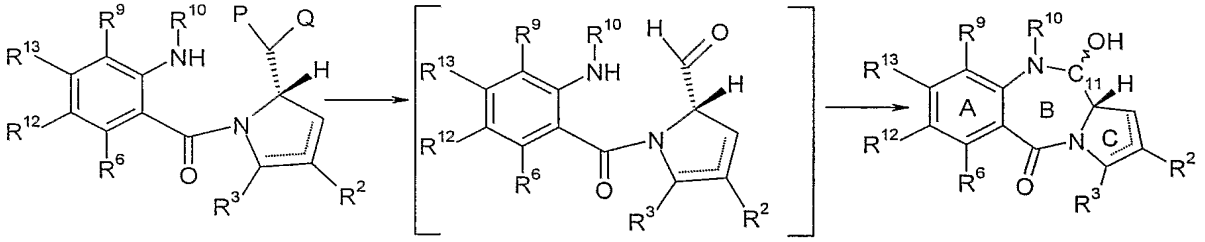
In general any nuceleophilic solvent is capable of forming such solvates as illustrated above for hydoxylic solvents. Other nuceleophilic solvents include thiols and amines.

These solvates may be isolated in solid form, for example, by lyophilisation.

General synthetic routes

The synthesis of PBD compounds is extensively discussed in WO 00/12508, which discussion is incorporated herein by reference.

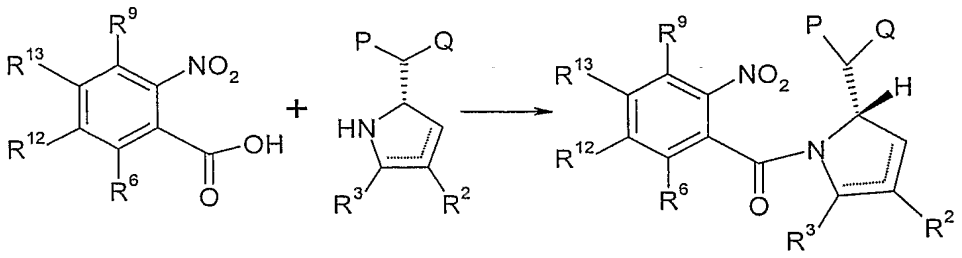
As discussed in that patent application, a key step in a preferred route to PBDs is a cyclisation to produce the B-ring, involving generation of an aldehyde (or functional equivalent thereof) at what will be the 11-position, and attack thereon by the Pro-N10-nitrogen:



IV

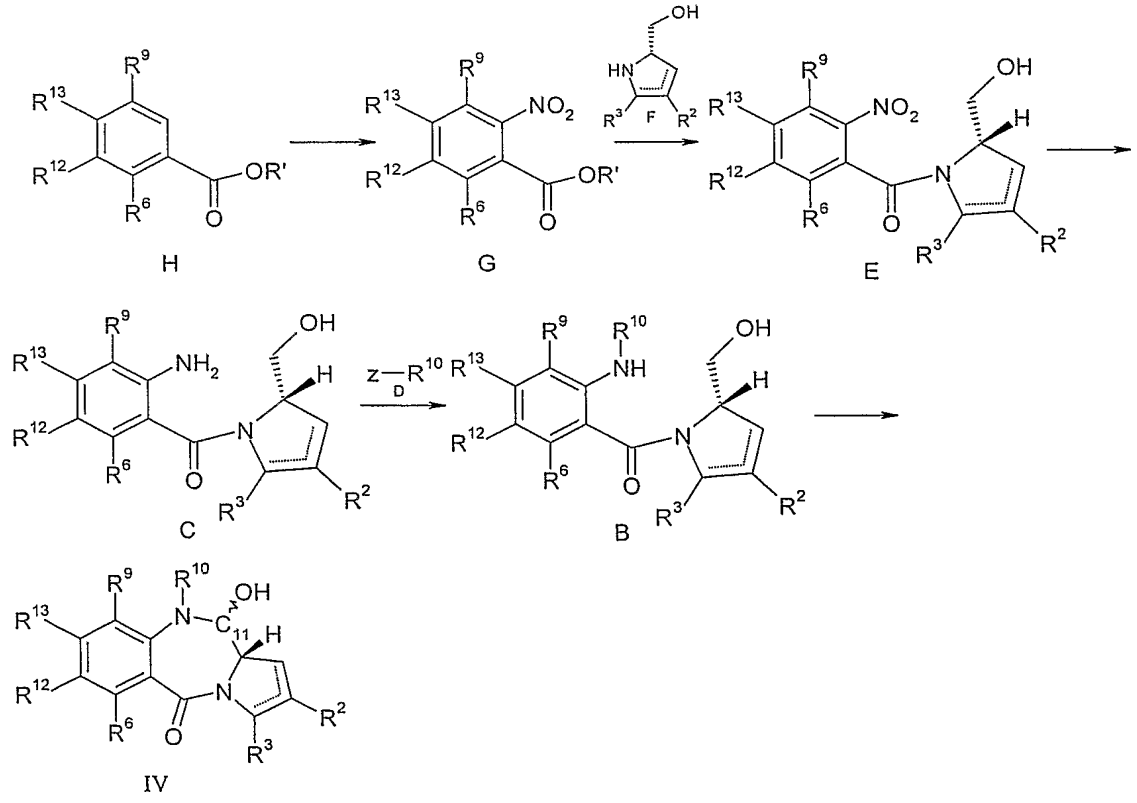
wherein the substituents are as defined in the second aspect of the invention and R^{12} and R^{13} are either OR^{14} and R^A respectively or R^A and OR^{14} repectively i.e. the protected hydroxyl group may be at either the C7 or C8 position with the other position being R^A. The "masked aldehyde" -CPQ may be an acetal or thioacetal, in which case the cyclisation involves unmasking. Alternatively, it may be an alcohol -CHOH, in which case the reaction involves oxidation, e.g. by means of TPAP, TEMPO or DMSO (Swern oxidation).

The masked aldehyde compound can be produced by condensing a corresponding 2,4-substituted pyrrolidine with a 2-nitrobenzoic acid:



The nitro group can then be reduced to $-NH_2$ and protected by reaction with a suitable agent, e.g. a chloroformate, which provides the removable nitrogen protecting group in the compound of formula **IV**.

A process involving the oxidation-cyclization procedure is illustrated in scheme 1 (an alternative type of cyclisation will be described later with reference to scheme 2).



Scheme 1

Exposure of the alcohol (B) (in which the Pro-N10-nitrogen is generally protected as carbamate) to tetrapropylammonium

perruthenate (TPAP)/N-methylmorpholine N-oxide (NMO) over A4 sieves results in oxidation accompanied by spontaneous B-ring closure to afford the desired product **IV**. The TPAP/NMO oxidation procedure is found to be particularly convenient for small scale reactions while the use of DMSO-based oxidation methods, particularly Swern oxidation, proves superior for larger scale work (e.g. > 1 g). A particularly preferred oxidising agent is (diacetoxyiodo)benzene (1.1 eq) and TEMPO (0.1 eq) dissolved in CH_2Cl_2.

The uncyclized alcohol (B) may be prepared by the reaction of a nitrogen protection reagent of formula D, which is preferably a chloroformate or acid chloride, to a solution of the amino alcohol C, generally in solution, generally in the presence of a base such as pyridine (preferably 2 equivalents) at a moderate temperature (e.g. at 0°C). Under these conditions little or no O-acylation is usually observed.

The key amino alcohol C may be prepared by reduction of the corresponding nitro compound E, by choosing a method which will leave the rest of the molecule intact. Treatment of E with tin (II) chloride in a suitable solvent, e.g. refluxing methanol, generally affords, after the removal of the tin salts, the desired product in high yield.

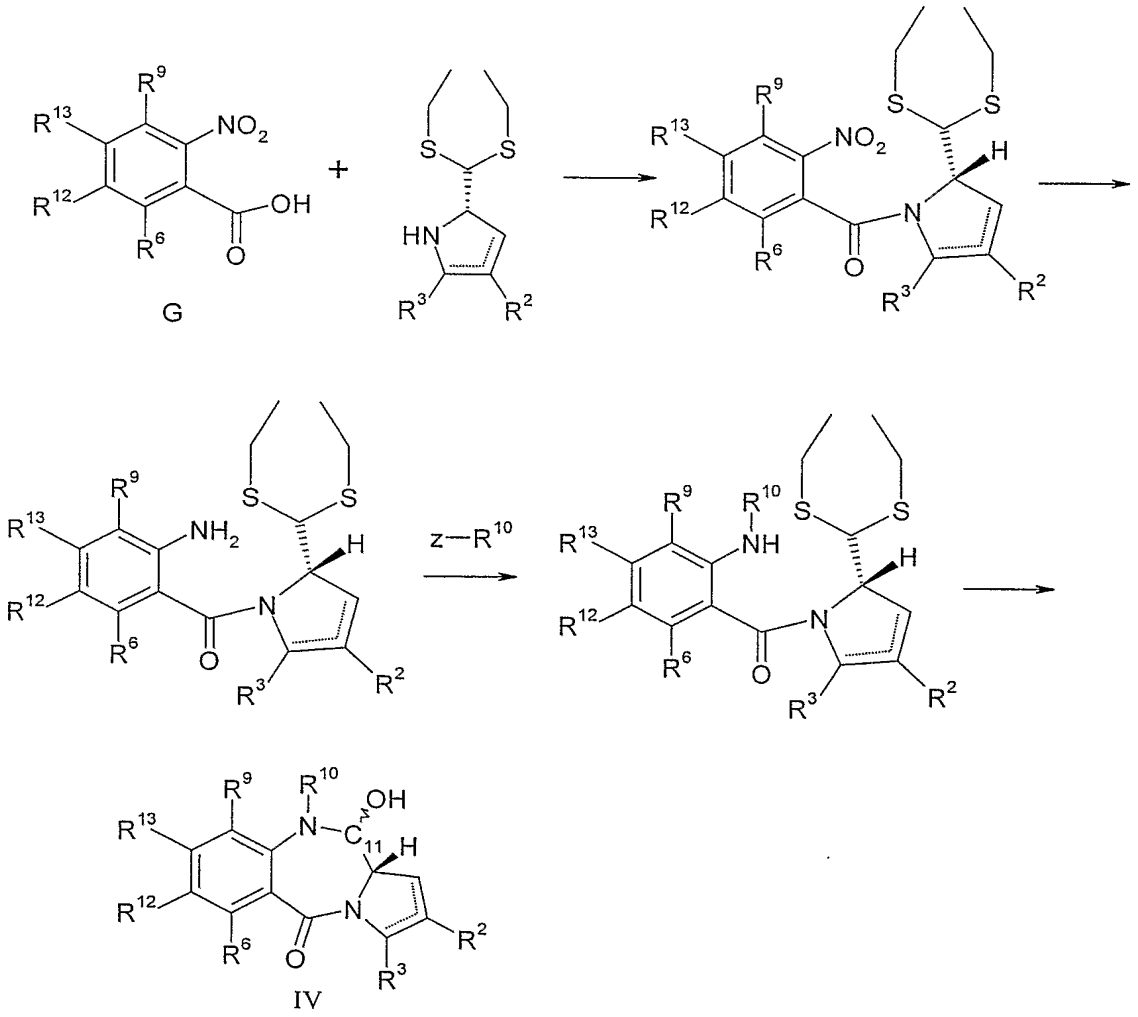
Exposure of E to hydrazine/Raney nickel avoids the production of tin salts and may result in a higher yield of C, although this method is less compatible with the range of possible C and A-ring substituents. For instance, if there is C-ring unsaturation (either in the ring itself, or in R_2 or R_3), this technique may be unsuitable.

The nitro compound of formula E may be prepared by coupling the appropriate *o*-nitrobenzoyl chloride to a compound of formula F, e.g. in the presence of K_2CO_3 at -25°C under a N_2 atmosphere. Compounds of formula F can be readily prepared, for example by

olefination of the ketone derived from L-trans-hydroxy proline. The ketone intermediate can also be exploited by conversion to the enol triflate for use in palladium mediated coupling reactions.

The *o*-nitrobenzoyl chloride is synthesised from the o-nitrobenzoic acid (or alkyl ester after hydrolysis) of formula G, which itself is prepared from the vanillic acid (or alkyl ester) derivative H. Many of these are commercially available and some are disclosed in Althuis, T.H. and Hess, H.J., *J. Medicinal Chem.*, **20(1)**, 146-266 (1977).

Alternative Cyclisation (Scheme 2)



Scheme 2

In scheme 1, the final or penultimate step was an oxidative cyclisation. An alternative, using thioacetal coupling, is shown in scheme 2. Mercury-mediated unmasking causes cyclisation to the protected PBD compound **IV**.

The thioacetal compound may be prepared as shown in scheme 2: the thioacetal protected C-ring [prepared via a literature method: Langley, D.R. & Thurston, D.E., *J. Organic Chemistry*, 52, 91-97 (1987)] is coupled to the o-nitrobenzoic acid (or alkyl ester after hydrolysis) (G) using a literature procedure. The resulting nitro compound cannot be reduced by hydrogenation, because of the

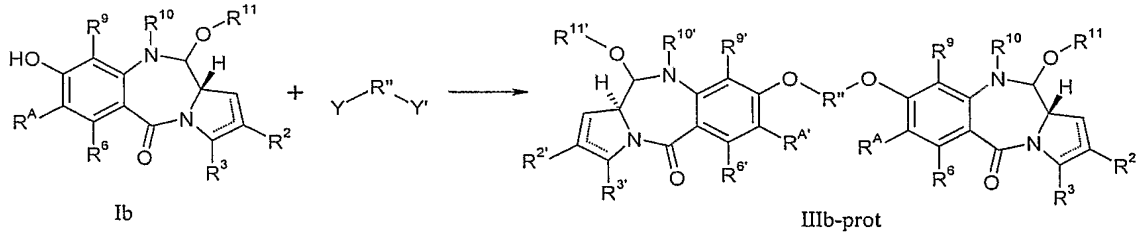
thioacetal group, so the tin(II) chloride method is used to afford the amine. This is then N-protected, e.g., by reaction with a chloroformate or acid chloride, such as 2,2,2-trichloroethylchloroformate.

Acetal-containing C-rings can be used as an alternative in this type of route with deprotection involving other methods, including the use of acidic conditions.

Alternative routes to PBDs

Alternative methods of synthesising N10 protected PBDs are disclosed in co-pending application GB0321295.8 (filed 11 September 2003), which describes the use of isocyanate intermediates.

*Formation of compounds **IIIa** and **IIIb** (Scheme 3)*



Scheme 3

Formation of the protected compound **IIIb** dimer from compound **Ib** is illustrated, however, protected compound **IIIa** is formed in an analogous manner with compound **Ia** as the starting material.

The PBD dimer compound **IIIa** or **IIIb** may be synthesized by dimerisation of compounds of formula **Ia** or **Ib** respectively following deprotection of the OH group at either the C7 or C8 position. The synthesis route illustrated in scheme 3 shows compounds when the dimer linkage is of the formula $-O-(CH_2)_n-O-$.

The protected dimer **IIIa** or **IIIb** may be formed from compounds of formula **Ia** or **Ib** respectively through reaction with a

disubstituted linking chain. The linking chain is preferably of the general form Y-R"-Y' where R" is as previously defined and Y and Y' are groups which can be reacted with an alcohol to form an ether linkage. Y and Y' are preferably independently selected from I, Br, Cl, OH, mesylate or tosylate. In a preferred aspect, Y and Y' are the same. In a preferred aspect Y and Y' are both iodo- groups.

Where Y and/or Y' is I, Br, Cl, mesylate or tosylate, the Y-R"-Y' reactant is coupled to the compound of formula **Ia** or **Ib** by a simple elimination reaction with Y and Y' as leaving groups. For example where the linking chain is $-O-CH_2-CH_2-CH_2-O-$, the compound of formula **Ia** or **Ib** is reacted with 1,3-diiodopropane in the presence of K_2CO_3. Generally, where the linking chain is a straight chain alkyl ether of the form $-O-(CH_2)_n-O-$, the compound of formula **Ia** or **Ib** is preferably reacted with the corresponding 1,n-diiodoalkane.

Where Y and/or Y' is OH, the Y-R"-Y' reactant is coupled to the compound of formula **Ia** or **Ib** under Mitsunobu conditions.

It is important that the OH protecting group at C11 in formula **Ia** or **Ib** is orthogonal to the OH protecting group at C7 and/or C8. This allows the C7 and/or C8 protection to be removed to give the free alcohol to allow dimerisation whilst the C11 OH group remains protected and therefore unreactive under the dimerisation conditions.

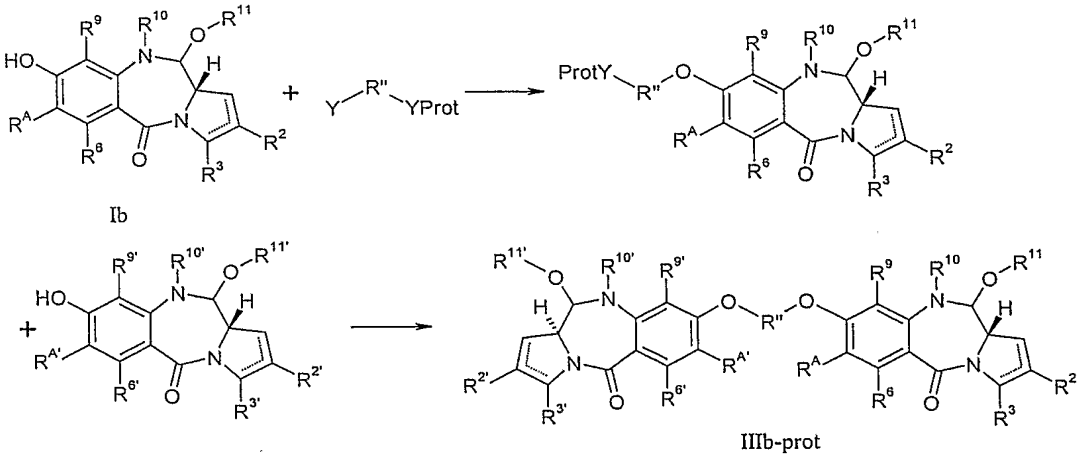
Following dimerisation, the imine bond in the compound of formula **IIIb-prot** can be deprotected by standard methods to yield the unprotected compound **IIIb** (which may be in its carbinolamine or carboinolamine ether form , depending on the solvents used). For example if R^{10} is Alloc, then the deprotection is carried using palladium to remove the N10 protecting group, followed by the elimination of water. If R^{10} is Troc, then the deprotection is

carried out using a Cd/Pb couple to yield the compound of formula **IIIb**.

Compound **IIIa** may be formed in an analogous manner via deprotection of the protected imine.

If the nitrogen protecting group (R^{10}) is such that the desired end product still contains it, e.g. if it is removable *in vivo*, then the C11 deprotected forms of compounds of formula **IIIa** or **IIIb** may be synthesised by removal of the oxygen protecting groups under suitable conditions to leave the R^{10} group in unaffected.

The above described methods are suited to the synthesis of dimers where both the PBD monomers have the same substituent pattern. One method of synthesising a dimer where the substituent pattern of the two PBD monomers is not the same involves protecting one end of the compound Y-R''-Y' (or using an already protected compound), coupling a PBD monomer to the unprotected end, deprotecting the other end and coupling a different PBD monomer to the free end. This route is shown in scheme 4.

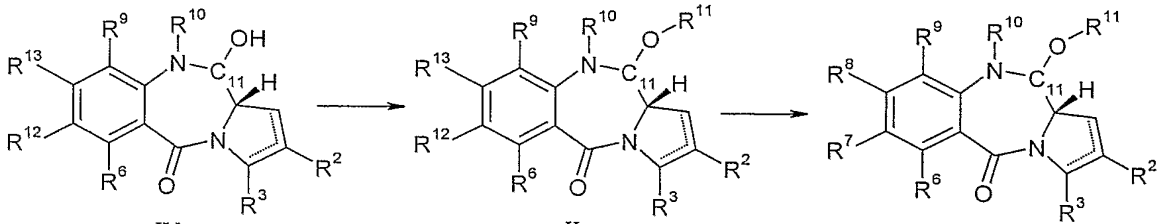


Scheme 4

Where Yprot, is a protected version, or precursor to Y'. If Y' is protected then the protecting group used should be orthogonal to those on the rest of the molecule, in particular, R^{10} and R^{11}. One example of this route, would be to have Y as -OH and YProt as -O-

benzyl. The first monomer could be joined by Mitsunobu coupling, the benzyl hydroxy deprotected, and then the free hydroxy coupled to the second monomer by a further Mitsunobu reaction.

*Formation of Compound of formula **Ia** or **Ib***



IV II I

Following cyclisation to form the B-ring, the C11-alcohol **IV** is then preferably protected, by conventional means to provide **II**. For example, if R^{11} is THP, the protection can take place by reacting **IV** with dihydropyran (DHP) and catalytic *p*-toluene sulfonic acid. Cleavage of the C7 or C8-protecting group from **II** then provides the corresponding C7 or C8 alcohol. For example, where the C7 or C8 protecting group (R^{12} or R^{13}) is a benzyl ether, this deprotection may be performed by reaction with H_2 catalysed by palladium on carbon.

This protection at the C11 position and deprotection of the C7 or C8 alcohol allows subsequent reaction of selectively the C7 or C8 alcohol position, for example to form the dimer compound **IIIa** or **IIIb** leaving the C11 position unaffected.

Further Preferences

The following preferences may apply to all aspects of the invention as described above, or may relate to a single aspect. The preferences may be combined together in any combination.

R^9 is preferably H.

R^2 is preferably R, and is more preferably an optionally substituted C_{5-20} aryl group. Most preferred is an optionally substituted phenyl group.

R^6 is preferably selected from H, OH, OR, SH, NH_2, nitro and halo, and is more preferably H or halo, and most preferably is H.

R^A is preferably independently selected from H, OR, SH, SR, NH_2, NHR, NHRR', and halo, and more preferably independently selected from H and OR, where R is preferably selected from optionally substituted C_{1-7} alkyl, C_{3-10} heterocyclyl and C_{5-10} aryl groups.

In the first aspect of the invention, R^{10} is preferably BOC or Troc. R^{11} is preferably THP or a silyl oxygen protecting group (for example TBS) and R^A is preferably selected from OMe and H. In a most preferred first aspect of the invention, R^{10} is BOC, R^{11} is THP and R^A is OMe.

In the second aspect of the invention, at least one of R^{14} is preferably a benzyl ether and R^A is preferably OMe or H. R^{11} is preferably THP or a silyl oxygen protecting group (for example TBS). Accordingly, in a particularly preferred embodiment of the second aspect of the invention R^A is OMe and R^{11} is THP or TBS. Furthermore, R^{10} is preferably BOC.

In some embodiments of the third aspect of the invention, R^{10} is preferably BOC and R^{15} is $O-R^{11}$, wherein R^{11} is preferably THP or a silyl oxygen protecting group (for example TBS).

In other embodiments of the third aspect of the invention, R^{10} and R^{15} together form a double bond between N10 and C11.

In some aspects of the third embodiment of the invention, the two PBD monomer units are linked at the C7 and C7' positions. In other aspects of the third embodiment of the invention, the two PBD monomer units are linked at the C8 and C8' positions.

In preferred aspects of the third embodiment of the invention, the substituent groups on C7, C8, N10 and C11 are the same on each

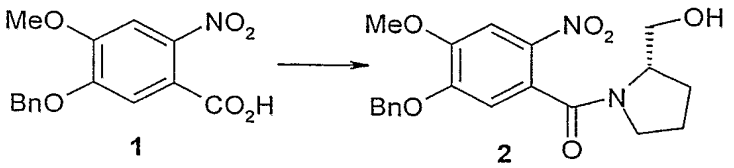
monomer unit that makes up the dimers of the third aspect of the invention. In is further preferred that the substituent groups on all positions of each mononmer unit that make up the dimer are the same.

Novel compounds of the present invention preferably have R^{10} and R^{15} forming a double bond between N10 and C11. Preferably, the novel compounds of the invention are dimers through C7 or C8, i.e. the R^7 or R^8 groups of each monomer form together a dimer bridge having the formula -X-R"-X- linking the monomers. More preferably, the dimer bridge is of formula $-O-(CH_2)_n-O-$, where n is 3 to 12, more preferably for the dimers linked at the C8 position, n is 7 to 12, more preferably n is 7 to 11 and even more preferably n is 7, 9 or 11; for the dimer linked at the C7 position, n is preferably 3 to 12, more preferably 3, 5, or 7. The preferences for R^6, R^7 and R^9 are as expressed above.

If R is optionally substituted C_{1-12} alkyl, it is preferred that it is optionally substituted C_{1-7} alkyl.

Example 1 - Synthesis of PBD monomer - (11*S*,11a*S*)-10-(*tert*-Butyloxycarbonyl)-7-hydroxy-8-methoxy-11-(tetrahydroxy-pyran-2-yloxy)-1,2,3,10,11,11a-hexahydro-5*H*-pyrrolo[2,1-c][1,4]benzodiazepine-5-one (7)

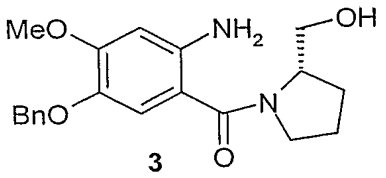
(a) (5-Benzyloxy-4-methoxy-2-nitrobenzoyl)-pyrrolidine-2-methanol (2)



1-(3-dimethylaminopropyl)-3-ethylcarbodiimide (EDCI) (6.33 g, 33.0 mmol, 1.0 equiv) was added to a stirred solution of the acid **1** (10 g, 33.0 mol, 1.0 equiv) in anhydrous DCM (300 mL) under a nitrogen atmosphere at 0°C. After stirring for 10 minutes the mixture was treated with HOBt (4.46 g, 33.0 mmol, 1.0 equiv) and few drops of

DMF and the resulting mixture was allowed to warm to room temperature and stirred for 2 h. The mixture was again cooled down to 0°C and treated with dropwise with a solution of pyrrolidinemethanol (5 g, 49.50 mmol, 1.5 equiv) in anhydrous DCM (100 mL). When the reaction mixture was complete, as indicated by TLC (EtOAc), the reaction mixture was diluted with DCM, washed with 1N HCl (100 mL), saturated $NaHCO_3$ (100 mL), brine (100 mL), dried ($MgSO_4$), filtered and evaporated *in vacuo*. The title compound was isolated by flash column chromatography (SiO_2, 50% EtOAc-hexane) to afford the coupled compound **2** (10 g, 25.7 mol, 78%) as a brown oil: $[\alpha]^{20}_D = -77°$ (c = 0.22, $CHCl_3$); ^1H NMR ($CDCl_3$, 400 MHz): δ 1.65-1.92 (m, 3H, 1-H, 2-H), 2.10-2.22 (m, 1H, 1-H), 2.96-3.08 (m, 2H, 3-H), 3.62-3.90 (m, 2H, 11-H), 3.98 (s, 3H, 7-OMe), 4.29-4.45 (m, 2H, 11a-H, OH), 5.24 (s, 2H, OBn), 6.81 (s, 1H, 6-H), 7.31- 7.47 (m, 5H, Ph), 7.72 (s, 1H, 9H); ^{13}C NMR ($CDCl_3$, 400 MHz): δ 24.3, 28.4, 49.4, 56.5, 61.4, 66.0, 71.4, 101.4, 110.9, 127.0, 127.1, 127.4, 128.3, 128.5, 128.81, 128.87, 135.0, 137.3, 149.6, 153.4; IR (neat): 3391, 2971, 2888, 1620, 1576, 1523, 1441, 1336, 1277, 1222, 1061 cm^{-1}; MS (FAB) *m/z* (relative intensity) 409 ($[M + Na]^{+\cdot}$, 58), 387 ($M^{+\cdot}$, 100), 285 (13).

(b) (2-Amino-5-benzyloxy-4-methoxybenzoyl)-pyrrolidine-2-methanol (3)

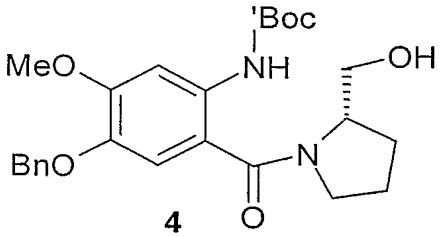


3

A solution of the nitro compound **2** (15.49 g, 40.12 mmol, 1.0 equiv) and tin (II) chloride (45.27 g, 200.64 mol, 5.0 equiv) in methanol (300 mL) was heated at reflux for 4 h. Excess solvent was removed by rotary evaporation under reduced pressure. The residue was treated carefully with a saturated aqueous sodium bicarbonate solution to basify the mixture to pH 9. The resulting suspension was allowed to stir overnight with ethyl acetate (100 mL), and filtrated through Celite to remove precipitated tin salts. The aqueous phase was extracted with EtOAc (2x50 mL), and

the combined organic phase washed with brine (50 mL), dried ($MgSO_4$), and evaporated *in vavuo* to provide **3** as a pink oil, which was used in the subsequent reaction without further purification: $[\alpha]^{20}_D$ = -97° (*c* = 0.18, $CHCl_3$); ^1H NMR ($CDCl_3$, 400·MHz): δ 1.50-1.81 (m, 3H, 1-H, 2-H), 2.05-2.15 (m, 1H, 1-H), 3.03-3.16 (m, 1H, 3-H), 3.23-3.36 (m, 1H, 3-H), 3.56-3.78 (m, 2H, 11-H), 3.88 (s, 3H, 7-OMe), 4.21-4.38 (m, 1H, 11a-H), 4.47-4.75 (m, 1H, 3-H), 5.03 (d, 1H, *J* = 12.4 Hz, OBn), 5.10 (d, 1H, *J* = 12.4 Hz, OBn), 6.26 (s, 1H, 9-H), 6.66 (s, 1H, 6-H), 7.28- 7.41 (m, 5H, Ph); ^{13}C NMR ($CDCl_3$, 400 MHz): δ 24.8, 28.5, 51.1, 55.7, 61.1, 67.5, 72.5, 100.8, 116.9, 127.5, 127.7, 128.4, 137.4, 138.8, 142.6, 153.0, 171.1, 171.7; IR (neat): 3436, 33.52, 29.67, 28.73, 1621, 1589, 1513, 1448, 1402, 1264, 1216, 1172, 1110, 1025cm^{-1}; MS (FAB) *m/z* (relative intensity) 379 ($[M + Na]^{+\cdot}$, 5), 357 ($M^{+\cdot}$, 100), 255 (58).

*(c) N-[5-Benzyloxy-2-(tert-butyloxycarbonylamino)-4-methoxybenzoyl]-pyrrolidine-2-methanol (**4**)*

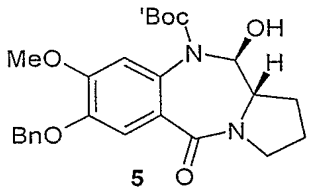


4

A solution of amine **3** (8 g, 22.47 mmol, 1.0 equiv) and Di-*tert*-butyl dicarbonate (7.35 g, 33.70 mmol, 1.5 equiv) in THF (150 mL) was heated at reflux overnight. The reaction mixture was allowed to cool to RT and excess THF was removed under reduced pressure to give the crude product. The residue was subjected to flash column chromatography (SiO_2, 30% EtOAc-hexane) to afford the product **4** (6.2 g, 13.59 mmol, 60%) as yellow oil: $[\alpha]^{20}_D$ = -106° (*c* = 0.198, $CHCl_3$); ^1H NMR ($CDCl_3$, 400 MHz): δ 1.47 (s, 9H, Boc),1.50-1.77 (m, 3H, 1-H, 2-H), 2.03-2.15 (m, 1H, 1-H), 2.83-3.00 (m, 1H, 3-H), 3.09-3.24 (m, 1H, 3-H), 3.56-3.84 (m, 2H, 11-H), 3.90 (s, 3H, 7-OMe), 4.21-4.43 (m, 2H, 11a-H, OH), 5.07 (d, 1H, *J* = 13 Hz, OBn), 5.19 (d, 1H, *J* = 13 Hz, OBn), 6.68 (s, 1H, 6-H), 7.26- 7.38 (m, 5H, Ph), 7.85 (s, 1H, 9-H), 8.60 (s, 1H, NH); ^{13}C NMR ($CDCl_3$, 400

MHz): δ 21.0, 28.33, 28.36, 51.1, 56.0, 61.1, 66.8, 71.6, 80.3, 104.4, 127.2, 127.8, 128.5, 133.5, 137.1, 141.4, 152.1, 153.1, 171.6; IR (neat): 3350, 2975, 1721, 1596, 1520, 1453, 1395, 1240, 1158, 1113, 1049 cm^{-1}; MS (FAB) *m/z* (relative intensity) 479 ([*M* + Na]$^{+\cdot}$, 40), 457 (*M* $^{+\cdot}$, 100), 357 (51).

(d) (11S,11aS)-7-Benzyloxy-10-(tert-butyloxycarbonyl)-11-hydroxy-8-methoxy-1,2,3,10,11,11a-hexahydro-5H-pyrrolo[2,1-c][1,4]benzodiazepine-5-one (5)

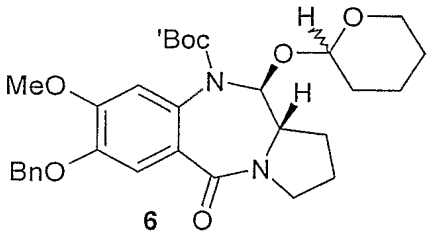


5

To a solution of Boc protected amine alcohol **4** (6.2 g, 13.59 mmol, 1.0 equiv) in DCM (50 mL), BAIB (4.82 g, 14.95 mmol, 1.1 equiv) and TEMPO (0.21 g, 1.35 mmol, 0.1 equiv) were added and the mixture was stirred overnight. When the reaction was complete as indicated by TLC (SiO_2, 50% EtOAc-hexane), the reaction mixture was diluted with DCM (100 mL) and washed with saturated $Na_2S_2O_3$ (60 mL). The aqueous layer was extracted with DCM (2x50 mL) and the combined organic layer was washed with brine (50 mL) and dried ($MgSO_4$). Removal of excess solvent under reduced pressure afforded a crude solid which was washed with cold EtOAc to give cyclized PBD **5** (4.9 g, 10.8 mmol, 79%) as white solid: $[\alpha]^{20}_D$ = +146° (*c* = 0.178, $CHCl_3$); ^{1}H NMR ($CDCl_3$, 400 MHz): δ 1.40 (s, 9H, Boc),1.93-2.18 (m, 4H, 1-H, 2-H), 3.42-3.50 (m, 1H, 11a-H), 3.51-3.61 (m, 1H, 3-H), 3.56-3.79 (m, 2H, 3-H, OH), 3.87 (s, 3H, 7-OMe), 5.13 (d, 1H, *J* = 12 Hz, OBn), 5.20 (d, 1H, *J* = 12 Hz, OBn), 5.51-5.62 (m, 1H, 11-H), 6.63 (s, 1H, 9-H), 7.29- 7.41 (m, 4H, 6-H, Ph), 7.43-7.48 (m, 2H, Ph); ^{13}C NMR ($CDCl_3$, 400 MHz): δ 23.0, 28.2, 28.7, 46.3, 56.1, 59.6, 70.9, 76.7, 77.0, 77.3, 81.7, 85.7, 112.3, 112.9, 125.4, 127.5, 128.0, 128.5, 129.6, 136.4, 147.2, 151.0, 159.0, 166.9; IR (neat): 3372, 2977, 2879, 1698, 1622, 1514, 1450, 1393, 1368, 1326, 1279, 1215, 1162, 1135, 1103, 1052, 1025cm^{-1}; MS

(FAB) *m/z* (relative intensity) 477 ([*M* + Na]$^{+\cdot}$, 35), 455 (*M* $^{+\cdot}$, 100), 399 (85), 337 (20).

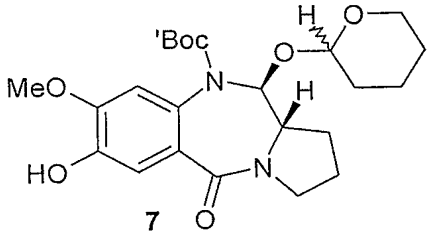
(e) (11S,11aS)-7-Benzyloxy-10-(tert-butyloxycarbonyl)-8-methoxy-11-(tetrahydroxy-pyran-2-yloxy)-1,2,3,10,11,11a-hexahydro-5H-pyrrolo[2,1-c][1,4]benzodiazepine-5-one (6)



6

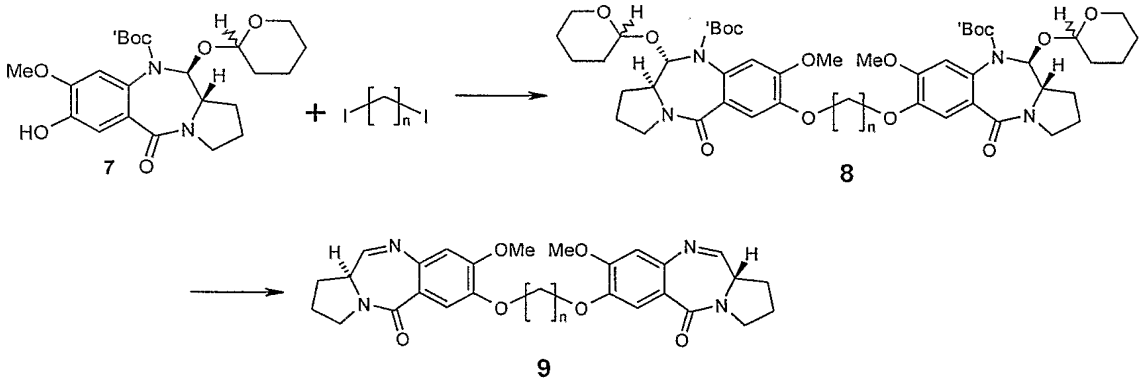
A catalytic amount of PTSA was added to a solution of DHP (2.87 g, 34.14 mmol, 5.0 equiv) in EtOAc (10 mL) at 0°C. After stirring 10 minutes, the cyclized compound **5** (3.1 g, 6.8 mmol, 1.0 equiv) was added portion-wise to the mixture and the resulting mixture was stirred until starting material disappearance by TLC (SiO_2, 50% EtOAc-hexane). The mixture was diluted with EtOAc (100 mL), washed with saturated $NaHCO_3$ (30mL), brine (30 mL) and dried ($MgSO_4$). Removal of excess solvent afforded the protected compound **6** (3.5 g, 6.5 mmol, 95% yield, mixture of diastereomers from THP protecting group), which was used in the subsequent reaction without further purification: $[\alpha]^{20}_D$ = +33° (*c* = 0.21, $CHCl_3$); ^{1}H NMR ($CDCl_3$, 400 MHz): δ 1.36 (s, 18H, Boc),1.47-1.92 (m, 12H, THP), 1.93-2.20 (m, 8H, 1-H, 2-H), 3.41-3.75 (m, 8H, 3-H, 11a-H, THP), 3.84-4.09 (m, 8H, 7-OMe, THP), 4.82-5.28 (m, 6H, OBn, THP), 5.69-5.79 (d, 1H, 11-H), 5.80-5.91 (d, 1H, 11-H), 6.54 (s, 1H, 9-H), 6.91 (s, 1H, 9-H), 7.27- 7.47 (m, 12H, 6-H, Ph); ^{13}C NMR ($CDCl_3$, 400 MHz): δ 19.8, 23.2, 25.31, 25.34, 25.4, 28.1, 28.2, 28.8, 29.1, 30.7, 30.9, 46.2, 55.9, 56.2, 60.0, 60.1, 63.3, 63.4, 70.94, 70.98, 81.0, 81.3, 88.2, 91.2, 98.4, 100.3, 112.0, 112.1, 113.6, 114.2, 126.4, 127.50, 127.54, 127.9, 128.0, 128.56, 128.58, 130.2, 136.5, 136.6, 147.4, 147.7, 151.0, 151.4, 159.0, 159.5, 167.2, 167.4; IR (neat): 3410, 2944, 2873, 1703, 1645, 1604, 1513, 1448, 1393, 1326, 1271, 1216, 1163, 1022cm^{-1}; MS (FAB) *m/z* (relative intensity) 561 ([*M* + Na]$^{+\cdot}$, 5), 539 (*M* $^{+\cdot}$, 100), 337 (82), 483 (24).

(f) (11S,11aS)-10-(tert-Butyloxycarbonyl)-7-hydroxy-8-methoxy-11-(tetrahydroxy-pyran-2-yloxy)-1,2,3,10,11,11a-hexahydro-5H-pyrrolo[2,1-c][1,4]benzodiazepine-5-one (7)



A catalytic amount of 10% palladium on carbon (380 mg) was added to a solution of THP protected compound **6** (3.8 g, 7 mmol) in absolute alcohol (30 mL). The reaction mixture was hydrogenated for 3h at 35 Psi. When the reaction was complete as indicated by TLC (SiO_2, 50%EtOAc-hexane) the reaction mixture was filtered through Celite, and removal of excess solvent under reduced pressure afforded the phenol **7** (2.8 g, 6.25 mmol, 90% yield, mixture of diastereomers from THP protecting group) as a white solid: $[\alpha]^{20}_D$ = +52° (*c* = 0.183, $CHCl_3$); ^1H NMR ($CDCl_3$, 400 MHz): δ 1.35 (s, 18H, Boc), 1.48-1.68 (m, 6H, THP), 1.69-1.88 (m, 6H, THP), 1.91-2.18 (m, 8H, 1-H, 2-H), 3.44-3.75 (m, 8H, 3-H, 11a-H, THP), 3.84-4.02 (m, 8H, 7-OMe, THP), 4.96-5.09 (m, 1H, THP), 5.10-5.18 (m, 1H, THP), 5.69-5.76 (d, 1H, 11-H), 5.77-5.87 (d, 1H, 11-H), 6.03 (s, 1H, OH), 6.14 (s, 1H, OH), 6.49 (s, 1H, 9-H), 6.86 (s, 1H, 9-H), 7.28 (s, 1H, 6-H), 7.32 (s, 1H, 6-H); ^{13}C NMR ($CDCl_3$, 400 MHz): δ 19.8, 20.5, 23.1, 23.2, 25.2, 25.3, 28.1, 28.2, 28.8, 29.1, 30.8, 31.2, 46.2, 55.9, 56.1, 59.9, 60.1, 63.3, 64.6, 80.9, 88.1, 91.1, 95.2, 100.4, 112.6, 113.3, 113.6, 114.2, 127.1, 129.0, 145.0, 145.3, 148.1, 148.5, 155.1, 167.1, 167.3; IR (neat): 3306, 2946, 1703, 1632, 1511, 1453, 1394, 1368, 1334, 1274, 1212, 1163, 1116, 1023cm^{-1}; MS (FAB) *m/z* (relative intensity) 471 ([*M* + Na]$^{+\cdot}$, 5), 449 (*M* $^{+\cdot}$, 100), 246 (50), 393 (22).

Examples 2-11 - Formation of PBD dimmers linked at the C-7 position (9)



n = 3,4,5,6,7,8,9,10,11,12

Example 2 (n=3)

*(a) 1,1'-[(Propane-1,3-diyl)dioxy]bis[(11S,11aS)-10-(tert-butyloxycarbonyl)-8-methoxy-11-(tetrahydro-pyran-2-yloxy)-1,2,3,10,11,11a-hexahydro-5H-pyrrolo[2,1-c][1,4]benzodiazepine-5-one] (**8a**)*

Diiodopropane (0.1 g, 0.22 mmol, 0.5 equiv) was added to the mixture of monomer **7** (0.2 g, 0.44 mmol, 1.0 equiv) and potassium carbonate (0.98 mmol, 2.2 equiv) in dry DMF (30 mL), and the resulting mixture was heated to 90°C under a nitrogen atmosphere for 5 hours. Removal of excess solvent under reduced pressure afforded a crude solid, which was subjected to flash column chromatography (SiO_2, 50%EtOAc-hexane) to afford the dimerized compound **8a** (80 mg, 0.08 mmol, 38% yield, mixture of diastereomers from THP protecting group as a solid: $[\alpha]^{20}_D$ = +31° (*c* = 0.16, $CHCl_3$); ^1H NMR ($CDCl_3$, 400 MHz): δ 1.35 (s, 36H, Boc), 1.44-1.67 (m, 16H, THP), 1.68-1.86 (m, 8H, THP), 1.91-2.20 (m, 16H, 1-H, 2-H), 2.35-2.44 (m, 4H, 13-H), 3.42-3.75 (m, 16H, 3-H, 11a-H, THP), 3.84-4.02 (m, 16H, 7-OMe, THP), 4.19-4.38 (m, 8H, 12-H), 5.01-5.10 (m, 2H, THP), 5.11-5.20 (m, 2H, THP), 5.69-5.77 (d, 2H, 11-H), 5.79-5.89 (d, 2H, 11-H), 6.50 (s, 2H, 9-H), 6.88 (s, 2H, 9-H), 7.22 (s, 2H, 6-H), 7.26 (s, 2H, 6-H); ^{13}C NMR ($CDCl_3$, 400 MHz): δ 19.8, 20.5, 23.1, 23.2, 25.3, 28.1, 28.2, 28.8, 29.0, 29.1, 30.9, 31.2, 46.3, 55.9, 56.1, 60.0, 60.1, 63.3, 64.5, 65.4, 81.0, 88.1, 91.2, 95.9, 100.2, 111.5, 113.5, 114.1, 129.9, 139.6, 144.5,

147.3, 147.8, 155.9, 167.4, 167.6; IR (neat): 3306, 2945, 1704, 1643, 1605, 1513, 1450, 1393, 1327, 1217, 1164, 1022cm^{-1}; MS (FAB) *m/z* (relative intensity) 937 ($M^{+\cdot}$, 100), 735 (25), 954 (14).

(b) 1,1′-[(Propane-1,3-diyl)dioxy]bis[(11aS)-8-methoxy-1,2,3,11a-tetrahydro-5H-pyrrolo[2,1-c][1,4]benzodiazepine-5-one] (9a)

95% TFA (3 mL) was added drop-wise to dimer compound **8a** (80 mg, 0.08 mmol) at 0°C. This was then stirred for 1hr and the mixture was poured into saturated $NaHCO_3$ (30 mL) solution to naturalize the reaction mixture. The mixture was extracted with chloroform (3x20 mL). The organic layer was then washed water (20 mL), brine (20 mL) then dried ($MgSO_4$) and filtrated. The excess solvent was removed under reduced pressure to give the crude product, which was subjected to flash column chromatography (SiO_2, 2%methanol-chloroform). Removal of excess eluent under reduced pressure without heating afforded the final product **9a** (31 mg, 0.06 mmol, 75%) as a solid: $[\alpha]^{20}_D$ = +515° (*c* = 0.10, $CHCl_3$); ^{1}H NMR ($CDCl_3$, 400 MHz): δ 2.01-2.11 (m, 4H, 2-H, 2′-H), 2.28-2.36 (m, 4H, 1-H, 1′-H), 2.38-2.45 (m, 2H, 13-H), 3.52-3.61 (m, 2H, 3-H, 3′-H), 3.67-3.75 (m, 2H, 11a-H, 11a′-H), 3.77-3.85 (m, 2H, 3-H, 3′-H), 3.90 (s, 6H, 7-OMe, 7′-OMe), 4.23-4.30 (m, 4H, 12-H, 12′-H), 6.79 (s, 2H, 9-H, 9′-H), 7.53 (s, 2H, 6-H, 6′-H), 7.65 (d, 2H, *J* = 4, 11-H, 11′-H); ^{13}C NMR ($CDCl_3$, 400 MHz): δ 24.1, 29.0, 29.6, 46.6, 53.7, 56.0, 65.6, 109.6, 112.9, 120.2, 140.7, 146.9, 151.8, 162.3, 164.6; IR (neat): 3350, 2956, 1599, 1506, 1447, 1385, 1262, 1216, 1091cm^{-1}; MS (FAB) *m/z* (relative intensity) 597 ($[M + 2 \times MeOH]^{+\cdot}$, 22), 565 ($[M + MeOH]^{+\cdot}$, 25), 533 ($M^{+\cdot}$, 100).

Example 3 (n=4)

(a) 1,1′-[(Butane-1,4-diyl)dioxy]bis[(11S,11aS)-10-(tert-butyloxycarbonyl)-8-methoxy-11-(tetrahydro-pyran-2-yloxy)-1,2,3,10,11,11a-hexahydro-5H-pyrrolo[2,1-c][1,4]benzodiazepine-5-one] (8b)

1,4-Diiodobutane (69.1 mg, was added to the mixture of monomer **7** (0.2 g, 0.44 mmol, 1.0 equiv) and potassium carbonate (0.98 mmol, 2.2 equiv) in dry DMF (30 mL), and the resulting mixture was

heated to 90°C under a nitrogen atmosphere for 5 h. Removal of excess solvent under reduced pressure afforded a crude solid, which was subjected to flash column chromatography (SiO_2, 50%EtOAc-hexane) to afford the dimerized compound **8b** (134 mg, 0.14 mmol, 63% yield, mixture of diastereomers from THP protecting group as a solid: $[\alpha]^{20}_D$ = +36° (*c* = 0.19, $CHCl_3$); ^1H NMR ($CDCl_3$, 400 MHz): δ 1.35 (s, 36H, Boc), 1.45-1.67 (m, 16H, THP), 1.68-1.86 (m, 8H, THP), 1.90-2.21 (m, 24H, 1-H, 2-H, 13-H), 3.44-3.78 (m, 16H, 3-H, 11a-H, THP), 3.84-4.02 (m, 16H, 7-OMe, THP), 4.04-4.25 (m, 8H, 12-H), 5.02-5.10 (m, 2H, THP), 5.11-5.20 (m, 2H, THP), 5.69-5.77 (d, 2H, 11-H), 5.79-5.89 (d, 2H, 11-H), 6.51 (s, 2H, 9-H), 6.88 (s, 2H, 9-H), 7.19 (s, 2H, 6-H), 7.28 (s, 2H, 6-H); ^{13}C NMR ($CDCl_3$, 400 MHz): δ 19.8, 20.5, 23.1, 23.2, 25.3, 25.9, 28.1, 28.2, 28.9, 29.1, 30.9, 31.2, 46.2, 55.9, 56.2, 60.1, 63.3, 63.6, 68.6, 80.9, 88.2, 91.2, 96.2, 100.2, 111.1, 111.4, 113.4, 114.1, 118.5, 126.4, 129.8, 143.1, 147.9, 148.2, 151.5, 151.8, 155.8, 167.4, 167.6; IR (neat): 2945, 1704, 1644, 1604, 1513, 1449, 1392, 1327, 1217, 1163, 1022cm^{-1}; MS (FAB) *m/z* (relative intensity) 973 ($[M + Na]^{+\cdot}$, 11), 951 ($M^{+\cdot}$, 100), 749 (36).

*(b) 1,1′-[(Butane-1,4-diyl)dioxy]bis[(11aS)-8-methoxy-1,2,3,11a-tetrahydro-5H-pyrrolo[2,1-c][1,4]benzodiazepine-5-one] (**9b**)*

95% TFA (3 mL) was added drop-wise to dimer compound **8b** (134 mg, 0.14 mmol) at 0°C. This was then stirred for 1hr and the mixture was poured into saturated $NaHCO_3$ (30 mL) solution to naturalize the reaction mixture. The mixture was extracted with chloroform (3x20 mL). The organic layer was then washed water (20 mL), brine (20 mL) then dried ($MgSO_4$) and filtrated. The excess solvent was removed under reduced pressure to give the crude product, which was subjected to flash column chromatography (SiO_2, 2%methanol-chloroform). Removal of excess eluent under reduced pressure without heating afforded the final product **9b** (60 mg, 0.11 mmol, 78%) as a solid: $[\alpha]^{20}_D$ = +477° (*c* = 0.09, $CHCl_3$); ^1H NMR ($CDCl_3$, 400 MHz): δ 1.88-2.06 (m, 8H, 2-H, 2′-H, 13-H, 13′-H), 2.16-2.36 (m, 4H, 1-H, 1′-H), 3.46-3.57 (m, 2H, 3-H, 3′-H), 3.63-3.69 (m, 2H, 11a-H, 11a′-H), 3.60-3.69 (m, 2H, 3-H, 3′-H), 3.82 (s, 6H, 7-OMe,

7'-OMe), 3.98-4.19 (m, 4H, 12-H, 12'-H), 6.72 (s, 2H, 9-H, 9'-H), 7.44 (s, 2H, 6-H, 6'-H), 7.59 (d, 2H, J = 4, 11-H, 11'-H); ^{13}C NMR ($CDCl_3$, 400 MHz): δ 24.1, 25.8, 29.65, 46.6, 53.7, 56.0, 68.6, 109.6, 112.6, 120.2, 140.6, 147.0, 151.7, 162.8, 164.6; IR (neat): 3354, 2950, 1622, 1600, 1506, 1447, 1387, 1262, 1216, 1092, 1026cm^{-1}; MS (FAB) *m/z* (relative intensity) 611 ([*M* + 2 x MeOH]$^{+\cdot}$, 9), 579 ([*M* + MeOH]$^{+\cdot}$, 19), 547 (*M* $^{+\cdot}$, 100).

Example 4 (n=5)

(a) 1,1'-[(Pentane-1,5-diyl)dioxy]bis[(11S,11aS)-10-(tert-butyloxycarbonyl)-8-methoxy-11-(tetrahydro-pyran-2-yloxy)-1,2,3,10,11,11a-hexahydro-5H-pyrrolo[2,1-c][1,4]benzodiazepine-5-one] (8c)

1,5-Diiodopentane (72.2 mg, 0.22 mmol, 0.5 equiv) was added to the mixture of monomer **7** (0.2 g, 0.44 mmol, 1.0 equiv) and potassium carbonate (0.98 mmol, 2.2 equiv) in dry DMF (30 mL), and the resulting mixture was heated to 90°C under a nitrogen atmosphere for 5 hours. Removal of excess solvent under reduced pressure afforded a crude solid, which was subjected to flash column chromatography (SiO_2, 50%EtOAc-hexane) to afford the dimerized compound **8c** (160 mg, 0.1 6mmol, 74% yield, mixture of diastereomers from THP protecting group) as a solid: $[\alpha]^{20}_D$ = +40° (*c* = 0.16, $CHCl_3$); ^{1}H NMR ($CDCl_3$, 400 MHz): δ 1.35 (s, 36H, Boc), 1.45-1.86 (m, 28H, 14-H, THP), 1.88-2.22 (m, 24H, 1-H, 2-H, 13-H), 3.44-3.77 (m, 16H, 3-H, 11a-H, THP), 3.82-4.02 (m, 16H, 7-OMe, THP), 4.03-4.19 (m, 8H, 12-H), 5.02-5.10 (m, 2H, THP), 5.11-5.20 (m, 2H, THP), 5.69-5.77 (d, 2H, 11-H), 5.79-5.89 (d, 2H, 11-H), 6.51 (s, 2H, 9-H), 6.88 (s, 2H, 9-H), 7.19 (s, 2H, 6-H), 7.22 (s, 2H, 6-H); ^{13}C NMR ($CDCl_3$, 400 MHz): δ 19.8, 20.4, 22.5, 23.1, 23.2, 25.3, 28.1, 28.2, 28.8, 29.1, 30.9, 31.2, 46.3, 55.9, 56.2, 60.0, 60.1, 63.3, 64.4, 68.8, 80.9, 81.2, 88.3, 91.4, 96.0, 100.4, 111.2, 113.5, 114.1, 126.5, 129.8, 135.8, 147.4, 148.0, 150.9, 154.4, 167.4, 167.6; IR (neat): 2945, 1704, 1643, 1604, 1513, 1449, 1392, 1327, 1217, 1163, 1022cm^{-1}; MS (FAB) *m/z* (relative intensity) 987 ([*M* + Na]$^{+\cdot}$, 14), 965 (*M* $^{+\cdot}$, 100), 863 (9).

*(b) 1,1'-[(Pentane-1,5-diyl)dioxy]bis[(11aS)-8-methoxy-1,2,3,11a-tetrahydro-5H-pyrrolo[2,1-c][1,4]benzodiazepine-5-one] (**9c**)*

95% TFA (3 mL) was added drop-wise to dimer compound **8c** (160 mg, 0.16 mmol) at 0°C. This was then stirred for 1hr and the mixture was poured into saturated $NaHCO_3$ (30 mL) solution to naturalize the reaction mixture. The mixture was extracted with chloroform (3x20 mL). The organic layer was then washed water (20 mL), brine (20 mL) then dried ($MgSO_4$) and filtrated. The excess solvent was removed under reduced pressure to give the crude product, which was subjected to flash column chromatography (SiO_2, 2%methanol-chloroform). Removal of excess eluent under reduced pressure without heating afforded the final product **9c** (72 mg, 0.13 mmol, 81 %) as a solid: $[\alpha]^{20}_D$ = +416° (*c* = 0.12, $CHCl_3$); ^1H NMR ($CDCl_3$, 400 MHz): δ 1.62-1.71 (m, 2H, 14-H), 1.88-1.99 (m, 4H, 13-H, 13'-H), 2.01-2.11 (m, 4H, 2-H, 2'-H), 2.26-2.36 (m, 4H, 1-H, 1'-H), 3.53-3.62 (m, 2H, 3-H, 3'-H), 3.69-3.75 (m, 2H, 11a-H, 11a'-H), 3.76-3.85 (m, 2H, 3-H, 3'-H), 3.90 (s, 6H, 7-OMe, 7'-OMe), 4.02-4.21 (m, 4H, 12-H, 12'-H), 6.79 (s, 2H, 9-H, 9'-H), 7.50 (s, 2H, 6-H, 6'-H), 7.65 (d, 2H, *J* = 4, 11-H, 11'-H); ^{13}C NMR ($CDCl_3$, 400 MHz): δ 22.5, 24.1, 28.8, 29.6, 46.6, 53.7, 56.0, 68.9, 109.6, 112.6, 120.2, 140.5, 147.0, 151.7, 162.3, 164.6; IR (neat): 3325, 2946, 1600, 1505, 1448, 1386, 1262, 1217, 1091, 1023cm^{-1}; MS (FAB) *m/z* (relative intensity) 625 ($[M + 2 \times MeOH]^{+\cdot}$, 19), 593 ($[M + MeOH]^{+\cdot}$, 25), 561 ($M^{+\cdot}$, 100).

Example 5 (n=6)

*(a) 1,1'-[(Hexane-1,6-diyl)dioxy]bis[(11S,11aS)-10-(tert-butyloxycarbonyl)-8-methoxy-11-(tetrahydro-pyran-2-yloxy)-1,2,3,10,11,11a-hexahydro-5H-pyrrolo[2,1-c][1,4]benzodiazepine-5-one] (**8d**)*

1,6-Diiodohexane (75.3 mg, 0.00 mmol, 0.5 equiv) was added to the mixture of monomer **7** (0.2 g, 0.44 mmol, 1.0 equiv) and potassium carbonate (0.98 mmol, 2.2 equiv) in dry DMF (30 mL), and the resulting mixture was heated to 90°C under a nitrogen atmosphere for 5 hours. Removal of excess solvent under reduced pressure afforded a crude solid, which was subjected to flash column

chromatography (SiO_2, 50%EtOAc-hexane) to afford the dimerized compound **8d** (174 mg, 0.17 mmol, 79% yield, mixture of diastereomers from THP protecting group) as a solid: $[\alpha]^{20}_D$ = +44° (*c* = 0.16, $CHCl_3$); ^1H NMR ($CDCl_3$, 400 MHz): δ 1.25-1.67 (m, 60H, 14-H, Boc, THP), 1.70-2.24 (m, 32H, 1-H, 2-H, 13-H, THP), 3.44-3.77 (m, 16H, 3-H, 11a-H, THP), 3.84-4.18 (m, 24H, 12-H, 7-OMe, THP), 5.02-5.10 (m, 2H, THP), 5.11-5.20 (m, 2H, THP), 5.69-5.77 (d, 2H, 11-H), 5.79-5.89 (d, 2H, 11-H), 6.51 (s, 2H, 9-H), 6.87 (s, 2H, 9-H), 7.18 (s, 2H, 6-H), 7.22 (s, 2H, 6-H); ^{13}C NMR ($CDCl_3$, 400 MHz): δ 19.8, 20.3, 23.1, 23.2, 25.3, 25.8, 28.1, 28.2, 28.9, 29.0, 29.1, 30.9, 31.2, 46.3, 55.9, 56.2, 60.0, 60.1, 63.3, 64.1, 68.9, 80.9, 81.2, 88.2, 91.1, 95.0, 100.5, 111.2, 111.7, 113.5, 114.1, 118.2, 127.1, 134.8, 147.7, 148.0, 155.2, 162.3, 163.0, 167.4, 167.6; IR (neat): 2943, 1703, 1644, 1604, 1513, 1449, 1392, 1327, 1217, 1163, 1022cm^{-1}; MS (FAB) *m/z* (relative intensity) 1001 ($[M + Na]^{+\cdot}$, 10), 979 ($M^{+\cdot}$, 100), 777 (24), 877 (12).

*(b) 1,1′-[(Hexane-1,6-diyl)dioxy]bis[(11aS)-8-methoxy-1,2,3,11a-tetrahydro-5H-pyrrolo[2,1-c][1,4]benzodiazepine-5-one] (**9d**)*

95% TFA (3 mL) was added drop-wise to dimer compound **8d** (174 mg, 0.17 mmol) at 0°C. This was then stirred for 1hr and the mixture was poured into saturated $NaHCO_3$ (30 mL) solution to naturalize the reaction mixture. The mixture was extracted with chloroform (3x20 mL). The organic layer was then washed water (20 mL), brine (20 mL) then dried ($MgSO_4$) and filtrated. The excess solvent was removed under reduced pressure to give the crude product, which was subjected to flash column chromatography (SiO_2, 2%methanol-chloroform). Removal of excess eluent under reduced pressure without heating afforded the final product **9d** (86 mg, 0.15 mmol, 88%) as a solid: $[\alpha]^{20}_D$ = +500° (*c* = 0.09, $CHCl_3$); ^1H NMR ($CDCl_3$, 400 MHz): δ 1.39-1.55 (m, 4H, 14-H, 14′-H), 1.72-1.89 (m, 4H, 13-H, 13′-H), 1.91-2.05 (m, 4H, 2-H, 2′-H), 2.17-2.31 (m, 4H, 1-H, 1′-H), 3.45-3.56 (m, 2H, 3-H, 3′-H), 3.62-3.69 (m, 2H, 11a-H, 11a′-H), 3.70-3.79 (m, 2H, 3-H, 3′-H), 3.83 (s, 6H, 7-OMe, 7′-OMe), 3.95-4.13 (m, 4H, 12-H, 12′-H), 6.73 (s, 2H, 9-H, 9′-H), 7.43 (s, 2H, 6-H, 6′-H), 7.58 (d, 2H, *J* = 4, 11-H, 11′-H); ^{13}C NMR ($CDCl_3$,

400 MHz): δ 24.1, 25.7, 29.0, 29.6, 46.6, 53.7, 56.0, 69.0, 109.6, 112.5, 120.2, 140.5, 147.1, 151.7, 162.8, 164.6; IR (neat): 3385, 2945, 1622, 1599, 1506, 1447, 1387, 1261, 1217, 1093cm^{-1}; MS (FAB) *m/z* (relative intensity) 639 ([*M* + 2 x MeOH]$^{+\cdot}$, 4), 607 ([*M* + MeOH]$^{+\cdot}$, 12), 575 (*M* $^{+\cdot}$, 100).

Example 6 (n=7)

*(a) 1,1'-[(Heptane-1,7-diyl)dioxy]bis[(11S,11aS)-10-(tert-butyloxycarbonyl)-8-methoxy-11-(tetrahydro-pyran-2-yloxy)-1,2,3,10,11,11a-hexahydro-5H-pyrrolo[2,1-c][1,4]benzodiazepine-5-one] (**8e**)*

1,7-Dibromoheptane (57.5 mg, 0.22 mmol, 0.5 equiv) was added to the mixture of monomer **7** (0.2 g, 0.44 mmol, 1.0 equiv) potassium carbonate (0.98 mmol, 2.2 equiv) and a catalytic amount of potassium iodide in dry DMF (30 mL), and the resulting mixture was heated to 90°C under a nitrogen atmosphere for 5 h. Removal of excess solvent under reduced pressure afforded a crude solid, which was subjected to flash column chromatography (SiO_2, 50%EtOAc-hexane) to afford the dimerized compound **8e** (190 mg, 0.19 mmol, 88% yield, mixture of diastereomers from THP protecting group) as a solid: $[\alpha]^{20}_D$ = +59° (*c* = 0.16, $CHCl_3$); ^{1}H NMR ($CDCl_3$, 400 MHz): δ 1.25-1.67 (m, 64H, 14-H, 15-H, Boc, THP), 1.68-1.92 (16H, 13-H, THP), 1.93-2.21 (m, 16H, 1-H, 2-H), 3.44-3.75 (m, 16H, 3-H, 11a-H, THP), 3.84-4.17 (m, 24H, 12-H, 7-OMe, THP), 5.02-5.10 (m, 2H, THP), 5.11-5.20 (m, 2H, THP), 5.69-5.77 (d, 2H, 11-H), 5.79-5.89 (d, 2H, 11-H), 6.51 (s, 2H, 9-H), 6.87 (s, 2H, 9-H), 7.19 (s, 2H, 6-H), 7.22 (s, 2H, 6-H); ^{13}C NMR ($CDCl_3$, 400 MHz): δ 19.8, 20.4, 23.1, 23.2, 25.3, 25.9, 28.1, 28.2, 28.9, 29.0, 29.1, 30.9, 31.2, 46.3, 55.9, 56.2, 60.0, 60.1, 63.3, 64.4, 69.0, 80.6, 80.9, 88.2, 91.2, 96.4, 100.2, 111.3, 111.9, 113.4, 114.0, 115.5, 116.1, 126.3, 129.7, 149.6, 149.9, 151.1, 155.5, 167.4, 167.6; IR (neat): 2942, 1704, 1643, 1604, 1514, 1450, 1392, 1327, 1218, 1164, 1022cm^{-1}; MS (FAB) *m/z* (relative intensity) 1015 ([*M* + Na]$^{+\cdot}$, 12), 993 (*M* $^{+\cdot}$, 100), 791 (23), 891 (9).

*(b) 1,1'-[(Heptane-1,7-diyl)dioxy]bis[(11aS)-8-methoxy-1,2,3,11a-tetrahydro-5H-pyrrolo[2,1-c][1,4]benzodiazepine-5-one] (**9e**)*

95% TFA (3 mL) was added drop-wise to dimer compound **8e** (195 mg, 0.19 mmol) at 0°C. This was then stirred for 1hr and the mixture was poured into saturated $NaHCO_3$ (30 mL) solution to naturalize the reaction mixture. The mixture was extracted with chloroform (3x20 mL). The organic layer was then washed water (20 mL), brine (20 mL) then dried ($MgSO_4$) and filtrated. The excess solvent was removed under reduced pressure to give the crude product, which was subjected to flash column chromatography (SiO_2, 2%methanol-chloroform). Removal of excess eluent under reduced pressure without heating afforded the final product **9e** (76 mg, 0.12 mmol, 68 %) as a solid: $[\alpha]^{20}_D$ = +473° (*c* = 0.14, $CHCl_3$); ^1H NMR ($CDCl_3$, 400 MHz): δ 1.41-1.55 (m, 6H, 14-H, 14'-H, 15-H), 1.85-1.94 (m, 4H, 13-H, 13'-H), 2.04-2.10 (m, 4H, 2-H, 2'-H), 2.28-2.38 (m, 4H, 1-H, 1'-H), 3.56-3.63 (m, 2H, 3-H, 3'-H), 3.72-3.76 (m, 2H, 11a-H, 11a'-H), 3.79-3.85 (m, 2H, 3-H, 3'-H), 3.92 (s, 6H, 7-OMe, 7'-OMe), 4.01-4.19 (m, 4H, 12-H, 12'-H), 6.79 (s, 2H, 9-H, 9'-H), 7.49 (s, 2H, 6-H, 6'-H), 7.64 (d, 2H, *J* = 4, 11-H, 11'-H); ^{13}C NMR ($CDCl_3$, 400 MHz): δ 24.1, 25.9, 29.0, 29.2, 29.6, 46.6, 53.7, 56.0, 69.1, 109.6, 112.5, 120.2, 140.5, 147.1, 151.6, 162.2, 164.6; IR (neat): 3325, 2935, 1600, 1506, 1448, 1388, 1261, 1217, 1022cm^{-1}; MS (FAB) *m/z* (relative intensity) 653 ($[M + 2 \times MeOH]^{+\cdot}$, 14), 621 ($[M + MeOH]^{+\cdot}$, 20), 589 ($M^{+\cdot}$, 100).

Example 7 (n=8)

*(a) 1,1'-[(Octane-1,8-diyl)dioxy]bis[(11S,11aS)-10-(tert-butyloxycarbonyl)-8-methoxy-11-(tetrahydro-pyran-2-yloxy)-1,2,3,10,11,11a-hexahydro-5H-pyrrolo[2,1-c][1,4]benzodiazepine-5-one] (**8f**)*

1,8-Diiodooctane (81.6 mg, 0.22 mmol, 0.5 equiv) was added to the mixture of monomer **7** (0.2 g, 0.44 mmol, 1.0 equiv) and potassium carbonate (0.98 mmol, 2.2 equiv) in dry DMF (30 mL), and the resulting mixture was heated to 90°C under a nitrogen atmosphere for 5 h. Removal of excess solvent under reduced pressure afforded a crude solid, which was subjected to flash column chromatography

(SiO_2, 50%EtOAc-hexane) to afford the dimerized compound **8f** (191 mg, 0.18 mmol, 85% yield, mixture of diastereomers from THP protecting group) as a solid: $[\alpha]^{20}_D$ = +50° (*c* = 0.15, $CHCl_3$); ^1H NMR ($CDCl_3$, 400 MHz): δ 1.27-1.66 (m, 68H, 14-H, 15-H, Boc, THP), 1.68-1.91 (16H, 3-H, THP), 1.93-2.20 (m, 16H, 1-H, 2-H), 3.45-3.75 (m, 16H, 3-H, 11a-H, THP), 3.83-4.14 (m, 24H, 12-H, 7-OMe, THP), 5.02-5.10 (m, 2H, THP), 5.11-5.20 (m, 2H, THP), 5.69-5.77 (d, 2H, 11-H), 5.79-5.89 (d, 2H, 11-H), 6.51 (s, 2H, 9-H), 6.87 (s, 2H, 9-H), 7.19 (s, 2H, 6-H), 7.22 (s, 2H, 6-H); ^{13}C NMR ($CDCl_3$, 400 MHz): δ 19.8, 20.4, 23.1, 23.2, 25.3, 25.9, 28.1, 28.2, 28.9, 29.0, 29.1, 29.3, 30.9, 31.2, 46.3, 55.9, 56.2, 60.0, 60.1, 63.3, 64.4, 69.0, 80.9, 81.2, 88.2, 91.2, 95.8, 100.2, 111.1, 111.7, 113.4, 114.0, 122.0, 126.4, 129.7, 138.0, 147.8, 148.1, 150.1, 150.5, 155.7, 167.4, 167.6; IR (neat): 2941, 1704, 1643, 1604, 1514, 1450, 1392, 1327, 1218, 1164, 1022cm^{-1}; MS (FAB) *m/z* (relative intensity) 1029 ($[M + Na]^{+\cdot}$, 30), 1007 ($M^{+\cdot}$, 100), 905 (15).

*(b) 1,1′-[(Octane-1,8-diyl)dioxy]bis[(11aS)-8-methoxy-1,2,3,11a-tetrahydro-5H-pyrrolo[2,1-c][1,4]benzodiazepine-5-one] (**9f**)*

95% TFA (3 mL) was added drop-wise to dimer compound **8f** (191 mg, 0.18 mmol) at 0°C. This was then stirred for 1hr and the mixture was poured into saturated $NaHCO_3$ (30 mL) solution to naturalize the reaction mixture. The mixture was extracted with chloroform (3x20 mL). The organic layer was then washed water (20 mL), brine (20 mL) then dried ($MgSO_4$) and filtrated. The excess solvent was removed under reduced pressure to give the crude product, which was subjected to flash column chromatography (SiO_2, 2%methanol-chloroform). Removal of excess eluent under reduced pressure without heating afforded the final product **9f** (106 mg, 0.17 mmol, 97%) as a solid: $[\alpha]^{20}_D$ = +467° (*c* = 0.14, $CHCl_3$); ^1H NMR ($CDCl_3$, 400 MHz): δ 1.34-1.53 (m, 8H, 14-H, 14′-H, 15-H, 15′-H), 1.78-1.91 (m, 4H, 13-H, 13′-H), 1.96-2.10 (m, 4H, 2-H, 2′-H), 2.25-2.38 (m, 4H, 1-H, 1′-H), 3.54-3.63 (m, 2H, 3-H, 3′-H), 3.70-3.76 (m, 2H, 11a-H, 11a′-H), 3.77-3.88 (m, 2H, 3-H, 3′-H), 3.90 (s, 6H, 7-OMe, 7′-OMe), 4.04-4.17 (m, 4H, 12-H, 12′-H), 6.81 (s, 2H, 9-H, 9′-H), 7.52 (s, 2H, 6-H, 6′-H), 7.67 (d, 2H, *J* = 4, 11-H, 11′-H); ^{13}C NMR

($CDCl_3$, 400 MHz): δ 24.1, 25.8, 29.0, 29.1, 29.6, 46.6, 53.7, 56.0, 69.0, 109.6, 112.5, 120.2, 140.5, 147.1, 151.6, 162.3, 164.6; IR (neat): 3326, 2937, 1599, 1506, 1448, 1387, 1262, 1217, 1092, 1023cm^{-1}; MS (FAB) *m/z* (relative intensity) 667 ([*M* + 2 x MeOH]$^{+\cdot}$, 7), 635 ([*M* + MeOH]$^{+\cdot}$, 15), 603 (*M* $^{+\cdot}$, 100).

Example 8 (n=9)

*(a) 1,1'-[(Nonane-1,9-diyl)dioxy]bis[(11S,11aS)-10-(tert-butyloxycarbonyl)-8-methoxy-11-(tetrahydro-pyran-2-yloxy)-1,2,3,10,11,11a-hexahydro-5H-pyrrolo[2,1-c][1,4]benzodiazepine-5-one] (**8g**)*

1,9-Dibromononane (63.7 mg, 0.22 mmol, 0.5 equiv) was added to the mixture of monomer **7** (0.2 g, 0.44 mmol, 1.0 equiv) potassium carbonate (0.98 mmol, 2.2 equiv) and a catalytic amount of potassium iodide in dry DMF (30mL), and the resulting mixture was heated to 90°C under a nitrogen atmosphere for 5 h. Removal of excess solvent under reduced pressure afforded a crude solid, which was subjected to flash column chromatography (SiO_2, 50%EtOAc-hexane) to afford the dimerized compound **8g** (181 mg, 0.17 mmol, 79% yield, mixture of diastereomers from THP protecting group) as a solid: $[\alpha]^{20}_D$ = +56° (*c* = 0.16, $CHCl_3$); ^{1}H NMR ($CDCl_3$, 400 MHz): δ 1.27-1.66 (m, 72H, 14-H, 15-H, 16-H, Boc, THP), 1.68-1.92 (16H, 13-H, THP), 1.93-2.20 (m, 16H, 1-H, 2-H), 3.45-3.75 (m, 16H, 3-H, 11a-H, THP), 3.83-4.14 (m, 24H, 12-H, 7-OMe, THP), 5.02-5.10 (m, 2H, THP), 5.12-5.19 (m, 2H, THP), 5.69-5.77 (d, 2H, 11-H), 5.79-5.89 (d, 2H, 11-H), 6.51 (s, 2H, 9-H), 6.87 (s, 2H, 9-H), 7.19 (s, 2H, 6-H), 7.22 (s, 2H, 6-H); ^{13}C NMR ($CDCl_3$, 400 MHz): δ 19.8, 20.4, 23.1, 23.2, 25.2, 25.3, 25.9, 28.1, 28.2, 28.9, 29.0, 29.1, 29.2, 29.3, 29.4, 30.9, 31.2, 46.2, 55.9, 56.2, 60.0, 60.1, 63.3, 64.4, 69.0, 69.1, 80.9, 81.2, 88.2, 91.2, 95.8, 100.2, 111.1, 111.5, 113.4, 114.0, 120.5, 129.6, 138.2, 147.8, 148.1, 151.2, 151.5, 161.9, 167.4, 167.6; IR (neat): 2938, 1703, 1643, 1604, 1513, 1449, 1392, 1327, 1217, 1163, 1022cm^{-1}; MS (FAB) *m/z* (relative intensity) 1043 ([*M* + Na]$^{+\cdot}$, 21), 1021 (*M* $^{+\cdot}$, 100), 819 (20), 919 (16).

*(b) 1,1'-[(Nonane-1,9-diyl)dioxy]bis[(11aS)-8-methoxy-1,2,3,11a-tetrahydro-5H-pyrrolo[2,1-c][1,4]benzodiazepine-5-one] (**9g**)*

95% TFA (3 mL) was added drop-wise to dimer compound **8g** (170 mg, 0.18 mmol) at 0°C. This was then stirred for 1hr and the mixture was poured into saturated $NaHCO_3$ (30 mL) solution to naturalize the reaction mixture. The mixture was extracted with chloroform (3x20 mL). The organic layer was then washed water (20 mL), brine (20 mL) then dried ($MgSO_4$) and filtrated. The excess solvent was removed under reduced pressure to give the crude product, which was subjected to flash column chromatography (SiO_2, 2%methanol-chloroform). Removal of excess eluent under reduced pressure without heating afforded the final product **9g** (93 mg, 0.15 mmol, 88%) as a solid: $[\alpha]^{20}_D$ = +547° (*c* = 0.13, $CHCl_3$); ^1H NMR ($CDCl_3$, 400 MHz): δ 1.31-1.53 (m, 10H, 14-H, 14'-H, 15-H, 15'-H, 16-H), 1.83-1.94 (m, 4H, 13-H, 13'-H), 2.00-2.12 (m, 4H, 2-H, 2'-H), 2.27-2.38 (m, 4H, 1-H, 1'-H), 3.54-3.64 (m, 2H, 3-H, 3'-H), 3.69-3.78 (m, 2H, 11a-H, 11a'-H), 3.79-3.87 (m, 2H, 3-H, 3'-H), 3.92 (s, 6H, 7-OMe, 7'-OMe), 4.01-4.19 (m, 4H, 12-H, 12'-H), 6.81 (s, 2H, 9-H, 9'-H), 7.52 (s, 2H, 6-H, 6'-H), 7.67 (d, 2H, *J* = 4, 11-H, 11'-H); ^{13}C NMR ($CDCl_3$, 400 MHz): δ 24.1, 25.9, 29.0, 29.3, 29.4, 29.6, 46.6, 53.7, 56.0, 69.1, 109.6, 112.5, 120.2, 140.5, 147.1, 151.6, 162.2, 164.6; IR (neat): 3325, 2933, 1600, 1507, 1448, 1388, 1261, 1217, 1092, 1024cm^{-1}; MS (FAB) *m/z* (relative intensity) 617 ($M^{+\cdot}$, 100), 635 (10), 785 (6).

Example 9 (n=10)

*(a) 1,1'-[(Decane-1,10-diyl)dioxy]bis[(11S,11aS)-10-(tert-butyloxycarbonyl)-8-methoxy-11-(tetrahydro-pyran-2-yloxy)-1,2,3,10,11,11a-hexahydro-5H-pyrrolo[2,1-c][1,4]benzodiazepine-5-one] (**8h**)*

1,10-Diiododecane (87.8 mg, 0.22 mmol, 0.5 equiv) was added to the mixture of monomer **7** (0.2 g, 0.44 mmol, 1.0 equiv) and potassium carbonate (0.98 mmol, 2.2 equiv) in dry DMF (30 mL), and the resulting mixture was heated to 90°C under a nitrogen atmosphere for 5 h. Removal of excess solvent under reduced pressure afforded a crude solid, which was subjected to flash column chromatography

(SiO_2, 50%EtOAc-hexane) to afford the dimerized compound **8h** (191 mg, 0.18mmol, 82% yield, mixture of diastereomers from THP protecting group) as a solid: $[\alpha]^{20}_D$ = +75° (*c* = 0.10, $CHCl_3$); ^1H NMR ($CDCl_3$, 400 MHz): δ 1.25-1.59 (m, 76H, 14-H, 15-H, 16-H, Boc, THP), 1.68-1.92 (16H, 13-H, THP), 1.93-2.20 (m, 16H, 1-H, 2-H), 3.45-3.75 (m, 16H, 3-H, 11a-H, THP), 3.83-4.14 (m, 24H, 12-H, 7-OMe, THP), 5.02-5.10 (m, 2H, THP), 5.12-5.19 (m, 2H, THP), 5.69-5.77 (d, 2H, 11-H), 5.79-5.89 (d, 2H, 11-H), 6.49 (s, 2H, 9-H), 6.86 (s, 2H, 9-H), 7.17 (s, 2H, 6-H), 7.21 (s, 2H, 6-H); ^{13}C NMR ($CDCl_3$, 400 MHz): δ 19.9, 20.4, 23.1, 23.2, 25.2, 25.3, 25.9, 28.1, 28.2, 28.9, 29.0, 29.1, 29.2, 29.3, 29.5, 30.9, 31.3, 46.3, 55.9, 56.2, 60.0, 60.1, 63.3, 69.1, 80.9, 81.2, 88.2, 91.2, 95.8, 100.2, 111.1, 111.5, 113.4, 114.0, 121.6, 126.4, 141.0, 143.1, 148.1, 148.4, 155.4, 167.4, 167.6; IR (neat): 2937, 1703, 1643, 1604, 1513, 1450, 1392, 1327, 1218, 1164, 1022cm^{-1}; MS (FAB) *m/z* (relative intensity) 1057 ($[M + Na]^{+\cdot}$, 34), 1035 ($M^{+\cdot}$, 100), 833 (26), 933 (25).

*(b) 1,1'-[(Decane-1,10-diyl)dioxy]bis[(11aS)-8-methoxy-1,2,3,11a-tetrahydro-5H-pyrrolo[2,1-c][1,4]benzodiazepine-5-one] (**9h**)*

95% TFA (3 mL) was added drop-wise to dimer compound **8h** (191 mg, 0.18 mmol) at 0°C. This was then stirred for 1hr and the mixture was poured into saturated $NaHCO_3$ (30 mL) solution to naturalize the reaction mixture. The mixture was extracted with chloroform (3x20 mL). The organic layer was then washed water (20 mL), brine (20 mL) then dried ($MgSO_4$) and filtrated. The excess solvent was removed under reduced pressure to give the crude product, which was subjected to flash column chromatography (SiO_2, 2%methanol-chloroform). Removal of excess eluent under reduced pressure without heating afforded the final product **9h** (103 mg, 0.16 mmol, 90%) as a solid: $[\alpha]^{20}_D$ = +387° (*c* = 0.17, $CHCl_3$); ^1H NMR ($CDCl_3$, 400 MHz): δ 1.25-1.50 (m, 12H, 14-H, 14'-H, 15-H, 15'-H, 16-H, 16'-H), 1.72-1.92 (m, 4H, 13-H, 13'-H), 1.98-2.09 (m, 4H, 2-H, 2'-H), 2.25-2.38 (m, 4H, 1-H, 1'-H), 3.52-3.62 (m, 2H, 3-H, 3'-H), 3.68-3.73 (m, 2H, 11a-H, 11a'-H), 3.76-3.85 (m, 2H, 3-H, 3'-H), 3.90 (s, 6H, 7-OMe, 7'-OMe), 3.95-4.19 (m, 4H, 12-H, 12'-H), 6.79 (s,

2H, 9-H, 9′-H), 7.50 (s, 2H, 6-H, 6′-H), 7.64 (d, 2H, J = 4, 11-H, 11′-H); ^{13}C NMR ($CDCl_3$, 400 MHz): δ 24.1, 25.9, 29.0, 29.3, 29.4, 29.6, 46.6, 53.7, 56.0, 69.7, 109.6, 112.5, 120.2, 140.5, 147.1, 151.6, 162.2, 164.6; IR (neat): 3325, 2931, 1600, 1506, 1448, 1388, 1262, 1217, 1092, 1024cm^{-1}; MS (FAB) *m/z* (relative intensity) 695 ([*M* + 2 x MeOH]$^{+\cdot}$, 14), 663 ([*M* + MeOH]$^{+\cdot}$, 20), 631 (*M* $^{+\cdot}$, 100).

Example 10 (n=11)

*(a) 1,1′-[(Undecane-1,11-diyl)dioxy]bis[(11S,11aS)-10-(tert-butyloxycarbonyl)-8-methoxy-11-(tetrahydro-pyran-2-yloxy)-1,2,3,10,11,11a-hexahydro-5H-pyrrolo[2,1-c][1,4]benzodiazepine-5-one] (**8i**)*

1,11-Dibromoundecane (70.0 mg, 0.22 mmol, 0.5 equiv) was added to the mixture of monomer **7** (0.2 g, 0.44 mmol, 1.0 equiv) potassium carbonate (0.98 mmol, 2.2 equiv) and a catalytic amount of potassium iodide in dry DMF (30 mL), and the resulting mixture was heated to 90°C under a nitrogen atmosphere for 5 h. Removal of excess solvent under reduced pressure afforded a crude solid, which was subjected to flash column chromatography (SiO_2, 50%EtOAc-hexane) to afford the dimerized compound **8i** (217 mg, 0.20 mmol, 94% yield, mixture of diastereomers from THP protecting group) as a solid: $[\alpha]^{20}_D$ = +52° (*c* = 0.17, $CHCl_3$); ^{1}H NMR ($CDCl_3$, 400 MHz): δ 1.27-1.60 (m, 80H, 14-H, 15-H, 16-H, 17-H, Boc, THP), 1.71-1.89 (16H, 13-H, THP), 1.93-2.20 (m, 16H, 1-H, 2-H), 3.45-3.75 (m, 16H, 3-H, 11a-H, THP), 3.83-4.14 (m, 24H, 12-H, 7-OMe, THP), 5.02-5.10 (m, 2H, THP), 5.12-5.19 (m, 2H, THP), 5.69-5.77 (d, 2H, 11-H), 5.79-5.89 (d, 2H, 11-H), 6.51 (s, 2H, 9-H), 6.87 (s, 2H, 9-H), 7.19 (s, 2H, 6-H), 7.22 (s, 2H, 6-H); ^{13}C NMR ($CDCl_3$, 400 MHz): δ 19.9, 20.4, 23.1, 23.2, 25.3, 25.9, 28.1, 28.2, 28.9, 29.0, 29.1, 29.2, 29.3, 29.5, 30.9, 31.2, 46.3, 55.9, 56.2, 60.0, 60.1, 63.3, 64.4, 69.1, 80.9, 81.2, 88.2, 91.2, 95.8, 100.2, 111.1, 111.5, 113.4, 114.0, 126.3, 129.6, 134.1, 138.8, 148.0, 148.4, 155.3, 155.6, 167.4, 167.6; IR (neat): 2935, 1704, 1643, 1604, 1513, 1449, 1392, 1327, 1218, 1164, 1022cm^{-1}; MS (FAB) *m/z* (relative intensity) 1071 ([*M* + Na]$^{+\cdot}$, 16), 1049 (*M* $^{+\cdot}$, 100), 947 (15), 847 (13).

*(b) 1,1'-[(Undecane-1,11-diyl)dioxy]bis[(11aS)-8-methoxy-1,2,3,11a-tetrahydro-5H-pyrrolo[2,1-c][1,4]benzodiazepine-5-one] (**9i**)*

95% TFA (3 mL) was added drop-wise to dimer compound **8i** (217 mg, 0.20 mmol) at 0°C. This was then stirred for 1hr and the mixture was poured into saturated $NaHCO_3$ (30 mL) solution to naturalize the reaction mixture. The mixture was extracted with chloroform (3x20 mL). The organic layer was then washed water (20 mL), brine (20 mL) then dried ($MgSO_4$) and filtrated. The excess solvent was removed under reduced pressure to give the crude product, which was subjected to flash column chromatography (SiO_2, 2%methanol-chloroform). Removal of excess eluent under reduced pressure without heating afforded the final product **9i** (113 mg, 0.17 mmol, 87%) as a solid: $[\alpha]^{20}_D$ = +401° (*c* = 0.19, $CHCl_3$); ^1H NMR ($CDCl_3$, 400 MHz): δ 1.26-1.52 (m, 14H, 14-H, 14'-H, 15-H, 15'-H, 16-H, 16'-H, 17-H), 1.84-1.92 (m, 4H, 13-H, 13'-H), 2.01-2.11 (m, 4H, 2-H, 2'-H), 2.27-2.36 (m, 4H, 1-H, 1'-H), 3.55-3.64 (m, 2H, 3-H, 3'-H), 3.70-3.76 (m, 2H, 11a-H, 11a'-H), 3.78-3.87 (m, 2H, 3-H, 3'-H), 3.92 (s, 6H, 7-OMe, 7'-OMe), 4.02-4.20 (m, 4H, 12-H, 12'-H), 6.81 (s, 2H, 9-H, 9'-H), 7.52 (s, 2H, 6-H, 6'-H), 7.67 (d, 2H, *J* = 4, 11-H, 11'-H); ^{13}C NMR ($CDCl_3$, 400 MHz): δ 24.1, 25.9, 29.0, 29.3, 29.5, 29.6, 46.6, 53.7, 56.0, 69.1, 109.6, 112.5, 120.2, 140.5, 147.1, 151.6, 162.2, 164.6; IR (neat): 3325, 2928, 1600, 1507, 1448, 1261, 1217, 1023cm^{-1}; MS (FAB) *m/z* (relative intensity) 709 ([*M* + 2 x MeOH]$^{+\cdot}$, 14), 677 ([*M* + MeOH]$^{+\cdot}$, 20), 645 (*M* $^{+\cdot}$, 100).

Example 11 (n=12)

*(a) 1,1'-[(Dodecane-1,12-diyl)dioxy]bis[(11S,11aS)-10-(tert-butyloxycarbonyl)-8-methoxy-11-(tetrahydro-pyran-2-yloxy)-1,2,3,10,11,11a-hexahydro-5H-pyrrolo[2,1-c][1,4]benzodiazepine-5-one] (**8j**)*

1,12-Dibromododecane (73.1 mg, 0.22 mmol, 0.5 equiv) was added to the mixture of monomer **7** (0.2 g, 0.44 mmol, 1.0 equiv) potassium carbonate (0.98 mmol, 2.2 equiv) and a catalytic amount of potassium iodide in dry DMF (30 mL), and the resulting mixture was

heated to 90°C under a nitrogen atmosphere for 5 h. Removal of excess solvent under reduced pressure afforded a crude solid, which was subjected to flash column chromatography (SiO_2, 50%EtOAc-hexane) to afford the dimerized compound **8j** (208 mg, 0.19 mmol, 87% yield, mixture of diastereomers from THP protecting group) as a solid: $[\alpha]^{20}_D$ = +50° (*c* = 0.20, $CHCl_3$); ^1H NMR ($CDCl_3$, 400 MHz): δ 1.26-1.68 (m, 84H, 14-H, 15-H, 16-H, 17-H, Boc, THP), 1.69-1.89 (16H, 13-H, THP), 1.93-2.20 (m, 16H, 1-H, 2-H), 3.44-3.75 (m, 16H, 3-H, 11a-H, THP), 3.83-4.14 (m, 24H, 12-H, 7-OMe, THP), 5.02-5.10 (m, 2H, THP), 5.12-5.19 (m, 2H, THP), 5.69-5.77 (d, 2H, 11-H), 5.79-5.89 (d, 2H, 11-H), 6.50 (s, 2H, 9-H), 6.87 (s, 2H, 9-H), 7.19 (s, 2H, 6-H), 7.22 (s, 2H, 6-H); ^{13}C NMR ($CDCl_3$, 400 MHz): δ 19.9, 20.4, 23.1, 23.2, 25.2, 25.3, 25.9, 28.1, 28.2, 28.9, 29.0, 29.1, 29.2, 29.3, 29.56, 29.58, 30.9, 31.2, 46.2, 55.9, 56.2, 60.0, 60.1, 63.3, 64.4, 69.12, 69.15, 80.9, 81.2, 88.2, 91.2, 95.8, 100.2, 111.1, 111.5, 113.4, 114.0, 126.3, 129.6, 134.1, 138.8, 147.8, 148.1, 151.5, 155.3, 167.4, 167.6; IR (neat): 2932, 1703, 1643, 1604, 1513, 1450, 1392, 1327, 1218, 1164, 1022cm^{-1}; MS (FAB) *m/z* (relative intensity) 1085 ($[M + Na]^{+\cdot}$, 28), 1063 ($M^{+\cdot}$, 100), 961 (17), 861 (13).

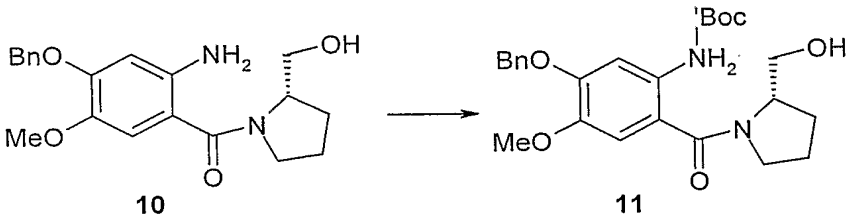
*(b) 1,1'-[(Dodecane-1,12-diyl)dioxy]bis[(11aS)-8-methoxy-1,2,3,11a-tetrahydro-5H-pyrrolo[2,1-c][1,4]benzodiazepine-5-one] (**9j**)*

95% TFA (3 mL) was added drop-wise to dimer compound **8j** (208 mg, 0.19 mmol) at 0°C. This was then stirred for 1hr and the mixture was poured into saturated $NaHCO_3$ (30 mL) solution to naturalize the reaction mixture. The mixture was extracted with chloroform (3x20 mL). The organic layer was then washed water (20 mL), brine (20 mL) then dried ($MgSO_4$) and filtrated. The excess solvent was removed under reduced pressure to give the crude product, which was subjected to flash column chromatography (SiO_2, 2%methanol-chloroform). Removal of excess eluent under reduced pressure without heating afforded the final product **9j** (107 mg, 0.16 mmol, 85%) as a solid: $[\alpha]^{20}_D$ = +506° (*c* = 0.15, $CHCl_3$); ^1H NMR ($CDCl_3$, 400 MHz): δ 1.26-1.52 (m, 16H, 14-H, 14'-H, 15-H, 15'-H, 16-H, 16'-H,

17-H, 17′-H), 1.83-1.92 (m, 4H, 13-H, 13′-H), 2.01-2.12 (m, 4H, 2-H, 2′-H), 2.28-2.37 (m, 4H, 1-H, 1′-H), 3.55-3.64 (m, 2H, 3-H, 3′-H), 3.70-3.77 (m, 2H, 11a-H, 11a′-H), 3.78-3.87 (m, 2H, 3-H, 3′-H), 3.92 (s, 6H, 7-OMe, 7′-OMe), 4.03-4.19 (m, 4H, 12-H, 12′-H), 6.81 (s, 2H, 9-H, 9′-H), 7.52 (s, 2H, 6-H, 6′-H), 7.67 (d, 2H, J = 4, 11-H, 11′-H); ^{13}C NMR ($CDCl_3$, 400 MHz): δ 24.1, 25.9, 29.0, 29.3, 29.54, 29.57, 29.6, 46.6, 53.7, 56.0, 69.1, 109.6, 112.5, 120.2, 140.5, 147.1, 151.6, 162.2, 164.6, ; IR (neat): 3338, 2926, 1600, 1507, 1448, 1261, 1217, 1024cm^{-1}; MS (FAB) *m/z* (relative intensity) 723 ([M + 2 x MeOH]$^{+\cdot}$, 14), 691 ([M + MeOH]$^{+\cdot}$, 20), 659 ($M^{+\cdot}$, 100).

Example 12 - Synthesis of PBD monomer - (11S,11aS)-8-Hydroxy-10-(tert-butyloxycarbonyl)-7-methoxy-11-(tetrahydroxy-pyran-2-yloxy)-1,2,3,10,11,11a-hexahydro-5H-pyrrolo[2,1-c][1,4]benzodiazepine-5-one (14)

*(a) N-[4-Benzyloxy-5-methoxy-2-(tert-butyloxycarbonylamino)benzoyl]-pyrrolidine-2-methanol (**11**)*

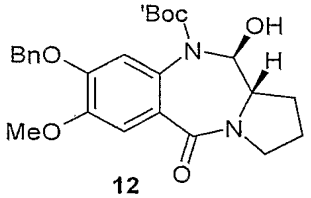


10 **11**

A solution of amine **10** (8 g, 22.47 mmol, 1.0 equiv) and Di-*tert*-butyl dicarbonate (7.35 g, 33.70 mmol, 1.5 equiv) in THF (150 mL) was heated at reflux overnight. The reaction mixture was allowed to cool to RT and excess THF was removed under reduced pressure to give the crude product. The residue was subjected to flash column chromatography (SiO_2, 30% EtOAc-hexane) to afford the product **11** (6.2 g, 13.59 mmol, 60%) as yellow oil: $[\alpha]^{20}_D$ = -95° (*c* = 0.17, $CHCl_3$); ^{1}H NMR ($CDCl_3$, 400 MHz): δ 1.50 (s, 9H, Boc), 1.64-1.80 (m, 2H, 1-H, 2-H), 1.86-1.95 (m, 1H, 2-H), 2.14-2.23 (m, 1H, 1-H), 3.46-3.54 (m, 1H, 3-H), 3.56-3.64 (m, 1H, 3-H), 3.67-3.77 (m, 1H, 11-H), 3.81-3.89 (m, 4H, 11-H, 7-OMe), 4.29-4.48 (m, 2H, 11a-H, OH), 5.14 (m, 2H, OBn), 6.82 (s, 1H, 6-H), 7.29- 7.33 (m, 1H, Ph), 7.34-7.39 (m, 2H, Ph), 7.46-7.49 (m, 2H, Ph), 7.89 (s, 1H, 9-H),

8.38 (br s, 1H, NH); ^{13}C NMR ($CDCl_3$, 400 MHz): δ 21.8, 28.3, 28.4, 52.4, 57.5, 61.8, 67.5, 71.5, 81.1, 107.0, 112.6, 128.3, 128.6, 128.8, 129.2, 133.2, 137.1, 144.7, 151.3, 153.9, 171.9; IR (neat): cm^{-1}; MS (FAB) *m/z* (relative intensity) 479 ([*M* + Na]$^{+\cdot}$, 20), 457 (*M* $^{+\cdot}$, 100), 357 (25), 255 (23), 401 (21).

*(b) (11S,11aS)-8-Benzyloxy-10-(tert-butyloxycarbonyl)-11-hydroxy-7-methoxy-1,2,3,10,11,11a-hexahydro-5H-pyrrolo[2,1-c][1,4]benzodiazepine-5-one (**12**)*

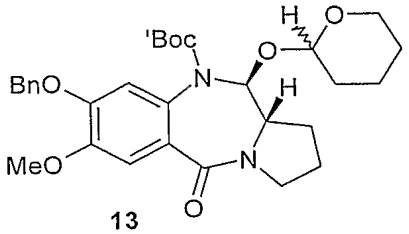


12

To a solution of Boc protected amine alcohol **11** (6.2 g, 13.59 mmol, 1.0 equiv) in DCM (50 mL), BAIB (4.82 g, 14.95 mmol, 1.0 equiv) and TEMPO (0.21 g, 1.35 mmol, 0.1 equiv) were added and the mixture was stirred overnight. When the reaction was complete as indicated by TLC (SiO_2, 50% EtOAc-hexane), the reaction mixture was diluted with DCM (100 mL) and washed with saturated $Na_2S_2O_3$ (60 mL). The aqueous layer was extracted with DCM (2x50 mL) and the combined organic layer was washed with brine (50 mL) and dried ($MgSO_4$). Removal of excess solvent under reduced pressure afforded a crude solid which was washed with cold EtOAc to give cyclized PBD **12** (4.9 g, 10.8 mmol, 79%) as white solid: $[\alpha]^{20}_D$ = +136° (*c* = 0.190, $CHCl_3$); ^{1}H NMR ($CDCl_3$, 400 MHz): δ 1.28 (s, 9H, Boc),1.95-2.18 (m, 4H, 1-H, 2-H), 3.43-3.50 (m, 1H, 11a-H), 3.53-3.66 (m, 2H, 3-H, OH), 3.67-3.78 (m, 1H, 3-H), 3.95 (s, 3H, 7-OMe), 5.10 (d, 1H, *J* = 12 Hz, OBn), 5.22 (d, 1H, *J* = 12 Hz, OBn), 5.51-5.59 (m, 1H, 11-H), 6.66 (s, 1H, 9-H), 7.26 (s, 1H, 6-H), 7.33- 7.34 (m, 1H, Ph), 7.38-7.41 (m, 2H, Ph), 7.41-7.45 (m, 2H, Ph); ^{13}C NMR ($CDCl_3$, 400 MHz): δ 23.0, 28.1, 28.7, 46.3, 56.1, 59.7, 71.1, 81.6, 85.6, 110.7, 114.6, 126.0, 127.0, 128.1, 128.7, 129.1, 130.2, 136.4, 148.5, 149.8, 155.4, 167.0; IR (neat): 3374, 2974, 1699, 1623, 1602, 1511, 1454, 1433, 1324, 1161, 1050cm^{-1}; MS (FAB) *m/z*

(relative intensity) 477 ([*M* + Na]$^{+\cdot}$, 25), 455 (*M* $^{+\cdot}$, 100), 399 (94), 337 (60), 437 (45).

(c) (11S,11aS)-8-Benzyloxy-10-(tert-butyloxycarbonyl)-7-methoxy-11-(tetrahydroxy-pyran-2-yloxy)-1,2,3,10,11,11a-hexahydro-5H-pyrrolo[2,1-c][1,4]benzodiazepine-5-one (13)

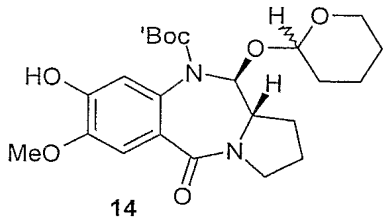


13

A catalytic amount of PTSA was added to a solution of DHP (2.87 g, 34.14 mmol, 5 equiv) in EtOAc (10 mL) at 0°C. After stirring 10 minutes, the cyclized compound **12** (3.1 g, 6.8 mmol, 1.0 equiv) was added portion-wise to the mixture and the resulting mixture was stirred until starting material disappearance by TLC (SiO_2, 50% EtOAc-hexane). The mixture was diluted with EtOAc (100 mL), washed with saturated $NaHCO_3$ (30 mL), brine (30 mL) and dried ($MgSO_4$). Removal of excess solvent afforded the protected compound **13** (3.5 g, 6.5 mmol, 95% yield, mixture of diastereomers from THP protecting group), which was used in the subsequent reaction without further purification: $[\alpha]^{20}_D$ = +64° (*c* = 0.58, $CHCl_3$); ^{1}H NMR ($CDCl_3$, 400 MHz): δ 1.12-1.80 (m, 30H, Boc, THP), 1.90-2.14 (m, 8H, 1-H, 2-H), 3.39-3.70 (m, 8H, 3-H, 11a-H, THP), 3.81-3.99 (m, 8H, 7-OMe, THP), 4.89-4.94 (m, 1H, THP), 5.05-5.26 (m, 5H, OBn, THP), 5.65-5.70 (d, 1H, 11-H), 5.74-5.81 (d, 1H, 11-H), 6.49 (s, 1H, 9-H), 6.88 (s, 1H, 9-H), 7.20- 7.36 (m, 12H, 6-H, Ph); ^{13}C NMR ($CDCl_3$, 400 MHz): δ 18.8, 19.6, 22.0, 22.2, 24.21, 24.25, 26.9, 27.0, 27.8, 28.0, 29.7, 30.2, 45.2, 55.06, 55.09, 58.95, 59.1, 62.2, 63.5, 69.9, 70.3, 79.8, 87.0, 90.2, 94.5, 99.5, 109.1, 109.6, 114.5, 114.7, 125.8, 126.93, 126.99, 127.65, 127.69, 128.7, 135.6, 135.7, 147.6, 147.9, 149.0, 149.1, 153.8, 166.3, 166.5; IR (neat): 3410, 2945, 2873, 1704, 1643, 1604, 1511, 1454, 1431, 1402, 1326, 1272, 1202, 1163, 1116, 1022cm^{-1}; MS (FAB) *m/z*

(relative intensity) 561 ([*M* + Na]$^{+\cdot}$, 57), 539 (*M* $^{+\cdot}$, 78), 337 (100), 540 (27), 338 (24).

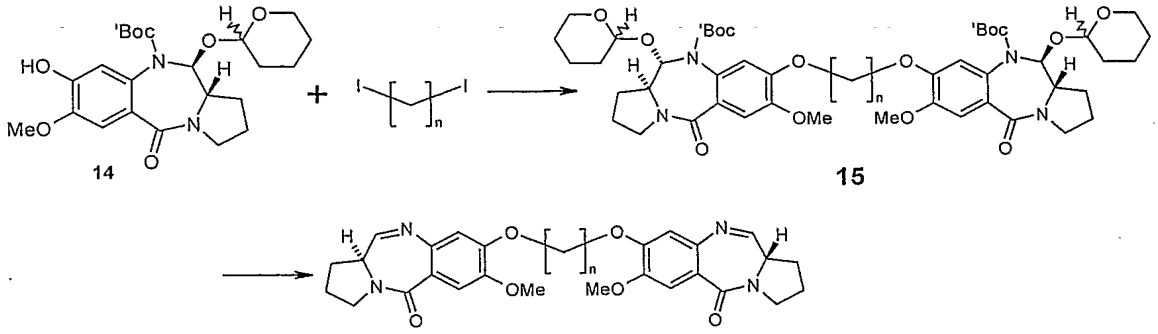
*(d) (11S,11aS)-8-Hydroxy-10-(tert-butyloxycarbonyl)-7-methoxy-11-(tetrahydroxy-pyran-2-yloxy)-1,2,3,10,11,11a-hexahydro-5H-pyrrolo[2,1-c][1,4]benzodiazepine-5-one (**14**)*



14

A catalytic amount of 10% palladium on carbon (380 mg) was added to a solution of THP protected compound **13** (3.8 g, 7 mmol) in absolute alcohol (30 mL). The reaction mixture was hydrogenated for 3h at 35 Psi. When the reaction was complete as indicated by TLC (SiO_2, 50%EtOAc-hexane) the reaction mixture was filtered through Celite, and removal of excess solvent under reduced pressure afforded the phenol **14** (2.8 g, 6.25 mmol, 90% yield, mixture of diastereomers from THP protecting group) as a white solid: $[\alpha]^{20}_D$ = +84° (*c* = 0.48, $CHCl_3$ ^{1}H NMR ($CDCl_3$, 400 MHz): δ 1.35 (s, 18H, Boc), 1.48-1.68 (m, 6H, THP), 1.69-1.88 (m, 6H, THP), 1.91-2.18 (m, 8H, 1-H, 2-H), 3.44-3.75 (m, 8H, 3-H, 11a-H, THP), 3.84-4.02 (m, 8H, 7-OMe, THP), 4.96-5.09 (m, 1H, THP), 5.10-5.18 (m, 1H, THP), 5.69-5.76 (d, 1H, 11-H), 5.77-5.87 (d, 1H, 11-H), 6.03 (s, 1H, OH), 6.14 (s, 1H, OH), 6.49 (s, 1H, 9-H), 6.86 (s, 1H, 9-H), 7.28 (s, 1H, 6-H), 7.32 (s, 1H, 6-H); ^{13}C NMR ($CDCl_3$, 400 MHz): δ 19.9, 20.7, 23.1, 23.2, 25.1, 25.3, 28.0, 28.1, 28.9, 29.1, 30.8, 31.2, 46.3, 56.13, 56.19, 60.0, 60.2, 63.4, 64.5, 81.0, 81.1, 87.9, 91.1, 95.8, 100.7, 109.6, 110.0, 116.4, 117.0, 125.5, 125.9, 130.2, 130.5, 145.7, 145.8, 147.4, 147.5, 154.9, 155.3, 167.4, 167.6; IR (neat): 3266, 2947, 1703, 1631, 1612, 1514, 1468, 1411, 1368, 1331, 1275, 1201, 1163, 1116, 1023cm^{-1}; MS (FAB) *m/z* (relative intensity) 471 ([*M* + Na]$^{+\cdot}$, 15), 449 (*M* $^{+\cdot}$, 99), 246 (100), 347 (25).

Examples 13-22: Synthesis of PBD dimers linked at the C-8 position



14 15 16

n = 3,4,5,6,7,8,9,10,11,12

Example 13 (n=3)

*(a) 1,1'-[(Propane-1,3-diyl)dioxy]bis[(11S,11aS)-10-(tert-butyloxycarbonyl)-7-methoxy-11-(tetrahydro-pyran-2-yloxy)-1,2,3,10,11,11a-hexahydro-5H-pyrrolo[2,1-c][1,4]benzodiazepine-5-one] (**15a**)*

1,3-Diiodopropane (66 mg, 0.22 mmol, 0.5 equiv) was added to the mixture of monomer **14** (0.2 g, 0.44 mmol, 1.0 equiv) and potassium carbonate (0.98mmol, 2.2 equiv) in dry DMF (30 mL), and the resulting mixture was heated to 90°C under a nitrogen atmosphere for 5 h. Removal of excess solvent under reduced pressure afforded a crude solid, which was subjected to flash column chromatography (SiO_2, 50%EtOAc-hexane) to afford the dimerized compound **15a** (90 mg, 0.09 mmol, 43% yield, mixture of diastereomers from THP protecting group) as a solid: $[\alpha]^{20}_D$ = +57° (*c* = 0.14, $CHCl_3$); ^1H NMR ($CDCl_3$, 400 MHz): δ 1.20-1.84 (m, 60H, Boc, THP), 1.91-2.20 (m, 16H, 1-H, 2-H), 2.34-2.46 (m, 4H, 13-H), 3.40-3.74 (m, 16H, 3-H, 11a-H, THP), 3.79-3.99 (m, 16H, 7-OMe, THP), 4.12-4.30 (m, 8H, 12-H), 4.97-5.15 (m, 4H, THP), 5.66-5.75 (d, 2H, 11-H), 5.77-5.89 (d, 2H, 11-H), 6.55 (s, 2H, 9-H), 6.89 (s, 2H, 9-H), 7.16 (s, 2H, 6-H), 7.20 (s, 2H, 6-H); ^{13}C NMR ($CDCl_3$, 400 MHz): δ 19.9, 20.5, 23.1, 23.2, 25.2, 25.3, 28.1, 28.2, 28.9, 29.0, 29.1, 30.9, 31.3, 46.3, 56.0, 56.1, 60.0, 60.2, 63.4, 64.5, 65.3, 65.7, 81.0, 81.1, 88.1, 91.2, 95.7, 100.3, 110.1, 110.8, 114.7, 115.2, 127.5, 129.8, 148.5, 148.8, 150.0, 155.1, 167.4, 167.6; IR (neat): 3426, 2943, 1703, 1643, 1604, 1513, 1454, 1432, 1326, 1270, 1201, 1163, 1023cm^{-}

^{1}; MS (FAB) *m/z* (relative intensity) 959 ([*M* + Na]$^{+\cdot}$, 100),937 (*M* $^{+\cdot}$, 62), 835 (67), 735 (60).

*(b) 1,1′-[(Propane-1,3-diyl)dioxy]bis[(11aS)-7-methoxy-1,2,3,11a-tetrahydro-5H-pyrrolo[2,1-c][1,4]benzodiazepine-5-one] (**16a**)*

95% TFA (3 mL) was added drop-wise to dimer compound **15a** (75 mg, 0.08 mmol) at 0°C. This was then stirred for 1h and the mixture was poured into saturated $NaHCO_3$ (30 mL) solution to naturalize the reaction mixture. The mixture was extracted with chloroform (3x20 mL). The organic layer was then washed water (20 mL), brine (20 mL) then dried ($MgSO_4$) and filtrated. The excess solvent was removed under reduced pressure to give the crude product, which was subjected to flash column chromatography (SiO_2, 2%methanol-chloroform). Removal of excess eluent under reduced pressure without heating afforded the final product **16a** (30 mg, 0.05 mmol, 70%) as a solid: $[\alpha]^{20}_D$ = +477° (*c* = 0.11, $CHCl_3$); ^{1}H NMR ($CDCl_3$, 400 MHz): δ 2.01-2.06 (m, 4H, 2-H, 2′-H), 2.28-2.31 (m, 4H, 1-H, 1′-H), 2.39-2.42 (m, 2H, 13-H), 3.55-3.60 (m, 2H, 3-H, 3′-H), 3.67-3.73 (m, 2H, 11a-H, 11a′-H), 3.77-3.87 (m, 2H, 3-H, 3′-H), 3.91 (s, 6H, 7-OMe, 7′-OMe), 4.23-4.31 (m, 4H, 12-H, 12′-H), 6.84 (s, 2H, 9-H, 9′-H), 7.50 (s, 2H, 6-H, 6′-H), 7.64 (d, 2H, *J* = 4, 11-H, 11′-H); ^{13}C NMR ($CDCl_3$, 400 MHz): δ 24.1, 28.4, 29.6, 46.6, 53.6, 56.1, 58.4, 65.4, 110.8, 111.6, 120.4, 140.6, 147.8, 150.6, 162.3, 164.6; IR (neat): 3350, 2951, 1600, 1505, 1434, 1262, 1217, 1021cm^{-1}; MS (FAB) *m/z* (relative intensity) 597 ([*M* + 2 x MeOH]$^{+\cdot}$, 16), 565 ([*M* + MeOH]$^{+\cdot}$, 5), 533 (*M* $^{+\cdot}$, 100).

Example 14 (n=4)

*(a) 1,1′-[(Butane-1,4-diyl)dioxy]bis[(11S,11aS)-10-(tert-butyloxycarbonyl)-7-methoxy-11-(tetrahydro-pyran-2-yloxy)-1,2,3,10,11,11a-hexahydro-5H-pyrrolo[2,1-c][1,4]benzodiazepine-5-one] (**15b**)*

1,4-Diiodobutane (69.1 mg, 0.22 mmol, 0.5 equiv) was added to the mixture of monomer **14** (0.2 g, 0.44mmol, 1.0 equiv) and potassium carbonate (0.98 mmol, 2.2 equiv) in dry DMF (30mL), and the resulting mixture was heated to 90°C under a nitrogen atmosphere

for 5 h. Removal of excess solvent under reduced pressure afforded a crude solid, which was subjected to flash column chromatography (SiO_2, 50%EtOAc-hexane) to afford the dimerized compound **15b** (210 mg, 0.22 mmol, 99% yield, mixture of diastereomers from THP protecting group) as a solid: $[\alpha]^{20}_D$ = -11° (*c* = 0.18, $CHCl_3$); ^1H NMR ($CDCl_3$, 400 MHz): δ 1.21-1.87 (m, 60H, Boc, THP), 1.94-2.21 (m, 24H, 1-H, 2-H, 13-H), 3.43-3.77 (m, 16H, 3-H, 11a-H, THP), 3.86-4.01 (m, 16H, 7-OMe, THP), 4.02-4.19 (m, 8H, 12-H), 4.97-5.15 (m, 4H, THP), 5.66-5.75 (d, 2H, 11-H), 5.77-5.89 (d, 2H, 11-H), 6.53 (s, 2H, 9-H), 6.88 (s, 2H, 9-H), 7.20 (s, 2H, 6-H), 7.24 (s, 2H, 6-H); ^{13}C NMR ($CDCl_3$, 400 MHz): δ 19.9, 20.6, 23.1, 23.2, 25.3, 25.9, 28.1, 28.2, 28.8, 29.1, 29.2, 30.9, 31.3, 46.3, 56.0, 56.1, 60.0, 60.1, 63.4, 64.6, 68.4, 68.8, 80.9, 81.2, 88.3, 91.2, 95.7, 100.4, 110.1, 110.7, 114.6, 115.1, 127.5, 129.8, 148.5, 148.8, 150.0, 155.0, 167.3, 167.6; IR (neat): 3472, 2945, 1704, 1643, 1604, 1513, 1454, 1432, 1327, 1271, 1202, 1163, 1023cm^{-1}; MS (FAB) *m/z* (relative intensity) 973 ([*M* + Na]$^{+\cdot}$, 46), 951 (*M* $^{+\cdot}$, 100), 968 (86), 849 (82), 749 (34).

*(b) 1,1'-[(Butane-1,4-diyl)dioxy]bis[(11aS)-7-methoxy-1,2,3,11a-tetrahydro-5H-pyrrolo[2,1-c][1,4]benzodiazepine-5-one] (**16b**)*

95% TFA (3 mL) was added drop-wise to dimer compound **15b** (170 mg, 0.17 mmol) at 0°C. This was then stirred for 1hr and the mixture was poured into saturated $NaHCO_3$ (30 mL) solution to naturalize the reaction mixture. The mixture was extracted with chloroform (3x20 mL). The organic layer was then washed water (20 mL), brine (20 mL) then dried ($MgSO_4$) and filtrated. The excess solvent was removed under reduced pressure to give the crude product, which was subjected to flash column chromatography (SiO_2, 2%methanol-chloroform). Removal of excess eluent under reduced pressure without heating afforded the final product **16b** (58 mg, 0.1 mmol, 62%) as a solid: $[\alpha]^{20}_D$ = +517° (*c* = 0.18, $CHCl_3$); ^1H NMR ($CDCl_3$, 400 MHz): δ 2.04-2.09 (m, 8H, 2-H, 2'-H, 13-H, 13'-H), 2.30-2.36 (m, 4H, 1-H, 1'-H), 3.57-3.62 (m, 2H, 3-H, 3'-H), 3.71-3.74 (m, 2H, 11a-H, 11a'-H), 3.79-3.85 (m, 2H, 3-H, 3'-H), 3.93 (s, 6H, 7-OMe, 7'-OMe), 4.11-4.21 (m, 4H, 12-H, 12'-H), 6.82 (s, 2H, 9-H, 9'-H),

7.52 (s, 2H, 6-H, 6′-H), 7.66 (d, 2H, J = 4, 11-H, 11′-H); ^{13}C NMR ($CDCl_3$, 400 MHz): δ 24.1, 25.7, 29.4, 46.6, 53.7, 56.1, 68.5, 110.5, 111.5, 120.2, 140.6, 147.8, 150.7, 162.3, 164.6; IR (neat): 3316, 2972, 1601, 1505, 1433, 1381, 1262, 1217, 1091cm^{-1}; MS (FAB) *m/z* (relative intensity) 611 ([*M* + 2 x MeOH]$^{+\cdot}$, 32), 579 ([*M* + MeOH]$^{+\cdot}$, 11), 547 (*M* $^{+\cdot}$, 100).

Example 15 (n=5)

*(a) 1,1′-[(Pentane-1,5-diyl)dioxy]bis[(11S,11aS)-10-(tert-butyloxycarbonyl)-7-methoxy-11-(tetrahydro-pyran-2-yloxy)-1,2,3,10,11,11a-hexahydro-5H-pyrrolo[2,1-c][1,4]benzodiazepine-5-one] (**15c**)*

1,5-Diiodopentane (72.2 mg, 0.22 mmol, 0.5 equiv) was added to the mixture of monomer **14** (0.2 g, 0.44 mmol, 1.0 equiv) and potassium carbonate (0.98 mmol, 2.2 equiv) in dry DMF (30 mL), and the resulting mixture was heated to 90°C under a nitrogen atmosphere for 5 h. Removal of excess solvent under reduced pressure afforded a crude solid, which was subjected to flash column chromatography (SiO_2, 50%EtOAc-hexane) to afford the dimerized compound **15c** (212 mg, 0.21 mmol, 98% yield, mixture of diastereomers from THP protecting group) as a solid: $[\alpha]^{20}_D$ = +40° (*c* = 0.22, $CHCl_3$); ^{1}H NMR ($CDCl_3$, 400 MHz): δ 1.21-1.83 (m, 64H, 14-H, Boc, THP), 1.88-2.18 (m, 24H, 1-H, 2-H, 13-H), 3.41-3.73 (m, 16H, 3-H, 11a-H, THP), 3.84-4.10 (m, 24H, 12-H, 7-OMe, THP), 4.97-5.15 (m, 4H, THP), 5.66-5.75 (d, 2H, 11-H), 5.77-5.89 (d, 2H, 11-H), 6.50(s, 2H, 9-H), 6.84 (s, 2H, 9-H), 7.17 (s, 2H, 6-H), 7.21 (s, 2H, 6-H); ^{13}C NMR ($CDCl_3$, 400 MHz): δ 19.9, 20.5, 21.0, 22.7, 23.1, 23.3, 25.3, 28.1, 28.2, 28.9, 29.1, 31.0, 31.3, 46.3, 56.0, 56.1, 60.0, 60.1, 63.4, 64.6, 68.7, 69.0, 80.9, 81.3, 88.2, 91.2, 95.7, 100.4, 110.2, 110.7, 114.5, 115.0, 126.3, 129.7, 129.8, 148.5, 148.8, 150.0, 155.1, 167.4, 167.6; IR (neat): 3431, 2945, 1704, 1643, 1604, 1513, 1453, 1432, 1327, 1271, 1202, 1163, 1023cm^{-1}; MS (FAB) *m/z* (relative intensity) 987 ([*M* + Na]$^{+\cdot}$, 37), 965 (*M* $^{+\cdot}$, 100), 763 (92), 863 (75), 982 (49).

*(b) 1,1'-[(Pentane-1,5-diyl)dioxy]bis[(11aS)-7-methoxy-1,2,3,11a-tetrahydro-5H-pyrrolo[2,1-c][1,4]benzodiazepine-5-one] (**16c**)*

95% TFA (3 mL) was added drop-wise to dimer compound **15c** (180 mg, 0.19 mmol) at 0°C. This was then stirred for 1hr and the mixture was poured into saturated $NaHCO_3$ (30 mL) solution to naturalize the reaction mixture. The mixture was extracted with chloroform (3x20 mL). The organic layer was then washed water (20 mL), brine (20 mL) then dried ($MgSO_4$) and filtrated. The excess solvent was removed under reduced pressure to give the crude product, which was subjected to flash column chromatography (SiO_2, 2%methanol-chloroform). Removal of excess eluent under reduced pressure without heating afforded the final product **16c** (70 mg, 0.12 mmol, 65%) as a solid: $[\alpha]^{20}_D$ = +626° (*c* = 0.17, $CHCl_3$); ^1H NMR ($CDCl_3$, 400 MHz): δ 1.62-1.71 (m, 2H, 14-H), 1.89-1.99 (m, 4H, 13-H, 13'-H), 2.0-2.11 (m, 4H, 2-H, 2'-H), 2.25-2.36 (m, 4H, 1-H, 1'-H), 3.53-3.62 (m, 2H, 3-H, 3'-H), 3.66-3.76 (m, 2H, 11a-H, 11a'-H), 3.77-3.85 (m, 2H, 3-H, 3'-H), 3.92 (s, 6H, 7-OMe, 7'-OMe), 4.01-4.16 (m, 4H, 12-H, 12'-H), 6.78 (s, 2H, 9-H, 9'-H), 7.50 (s, 2H, 6-H, 6'-H), 7.64 (d, 2H, *J* = 4, 11-H, 11'-H); ^{13}C NMR ($CDCl_3$, 400 MHz): δ 22.5, 29.6, 46.6, 53.7, 56.1, 68.7, 110.5, 111.6, 120.1, 140.6, 147.8, 150.8, 162.3, 164.6; IR (neat): 3350, 2946, 1600, 1505, 1455, 1433, 1383, 1262, 1217, 1092, 1020cm^{-1}; MS (FAB) *m/z* (relative intensity) 625 ([*M* + 2 x MeOH]$^{+\cdot}$, 4), 593 ([*M* + MeOH]$^{+\cdot}$, 12), 547 (*M* $^{+\cdot}$, 100).

Example 16 (n=6)

*(a) 1,1'-[(Hexane-1,6-diyl)dioxy]bis[(11S,11aS)-10-(tert-butyloxycarbonyl)-7-methoxy-11-(tetrahydro-pyran-2-yloxy)-1,2,3,10,11,11a-hexahydro-5H-pyrrolo[2,1-c][1,4]benzodiazepine-5-one] (**15d**)*

1,6-Diiodohexane(75.3 mg, 0.22 mg, 0.5 equiv) was added to the mixture of monomer **14** (0.2 g, 0.44 mmol, 1.0 equiv) and potassium carbonate (0.98 mmol, 2.2 equiv) in dry DMF (30 mL), and the resulting mixture was heated to 90°C under a nitrogen atmosphere for 5 h. Removal of excess solvent under reduced pressure afforded

a crude solid, which was subjected to flash column chromatography (SiO_2, 50%EtOAc-hexane) to afford the dimerized compound **15d** (190 mg, 0.19 mmol, 87% yield, mixture of diastereomers from THP protecting group) as a solid: $[\alpha]^{20}_D$ = +11° (*c* = 0.18, $CHCl_3$); ^1H NMR ($CDCl_3$, 400 MHz): δ 1.25-1.66 (m, 60H, 14-H, Boc, THP), 1.68-2.85 (m, 8H, THP), 1.86-2.20 (m, 24H, 1-H, 2-H, 13-H), 3.44-3.73 (m, 16H, 3-H, 11a-H, THP), 3.86-4.10 (m, 24H, 12-H, 7-OMe, THP), 5.01-5.17 (m, 4H, THP), 5.68-5.76 (d, 2H, 11-H), 5.78-5.89 (d, 2H, 11-H), 6.50(s, 2H, 9-H), 6.87 (s, 2H, 9-H), 7.19 (s, 2H, 6-H), 7.23 (s, 2H, 6-H); ^{13}C NMR ($CDCl_3$, 400 MHz): δ 19.8, 20.5, 23.1, 23.2, 25.3, 25.9, 28.1, 28.2, 28.9, 29.02, 29.06, 29.1, 29.2, 30.9, 31.2, 46.3, 56.1, 56.2, 60.0, 60.1, 63.3, 64.5, 68.8, 69.1, 80.9, 81.3, 88.2, 91.2, 95.7, 100.3, 110.1, 110.6, 114.5, 115.0, 126.3, 129.8, 148.5, 148.8, 150.0, 155.1, 167.4, 167.6; IR (neat): 2944, 1704, 1643, 1605, 1513, 1454, 1432, 1327, 1272, 1202, 1164, 1023cm^{-1}; MS (FAB) *m/z* (relative intensity) 1001 ([*M* + Na]$^{+\cdot}$, 21), 979 (*M*$^{+\cdot}$, 100), 877 (26), 777 (17).

*(b) 1,1′-[(Hexane-1,6-diyl)dioxy]bis[(11aS)-7-methoxy-1,2,3,11a-tetrahydro-5H-pyrrolo[2,1-c][1,4]benzodiazepine-5-one] (**16d**)*

95% TFA (3 mL) was added drop-wise to dimer compound **15d** (190 mg, 0.19 mmol) at 0°C. This was then stirred for 1hr and the mixture was poured into saturated $NaHCO_3$ (30 mL) solution to naturalize the reaction mixture. The mixture was extracted with chloroform (3x20 mL). The organic layer was then washed water (20 mL), brine (20 mL) then dried ($MgSO_4$) and filtrated. The excess solvent was removed under reduced pressure to give the crude product, which was subjected to flash column chromatography (SiO_2, 2%methanol-chloroform). Removal of excess eluent under reduced pressure without heating afforded the final product **16d** (99 mg, 0.17 mmol, 90%) as a solid: $[\alpha]^{20}_D$ = +474° (*c* = 0.19, $CHCl_3$); ^1H NMR ($CDCl_3$, 400 MHz): δ 1.49-1.65 (m, 4H, 14-H, 14′-H), 1.82-1.97 (m, 4H, 13-H, 13′-H), 1.99-2.13 (m, 4H, 2-H, 2′-H), 2.26-2.41 (m, 4H, 1-H, 1′-H), 3.53-3.65 (m, 2H, 3-H, 3′-H), 3.68-3.76 (m, 2H, 11a-H, 11a′-H), 3.77-3.89 (m, 2H, 3-H, 3′-H), 3.94 (s, 6H, 7-OMe, 7′-OMe), 4.01-4.17 (m, 4H, 12-H, 12′-H), 6.80 (s, 2H, 9-H, 9′-H), 7.52 (s,

2H, 6-H, 6'-H), 7.66 (d, 2H, J = 4, 11-H, 11'-H); ^{13}C NMR ($CDCl_3$, 400 MHz): δ 24.1, 25.7, 28.8, 29.6, 46.6, 53.7, 56.1, 68.8, 110.4, 111.6, 120.1, 140.6, 147.8, 150.8, 162.3, 164.6; IR (neat): 3318, 2943, 1601, 1506, 1454, 1433, 1382, 1262, 1217, 1021cm^{-1}; MS (FAB) *m/z* (relative intensity) 639 ([*M* + 2 x MeOH]$^{+\cdot}$, 6), 607 ([*M* + MeOH]$^{+\cdot}$, 23), 575 (*M* $^{+\cdot}$, 100).

Example 17 (n=7)

*(a) 1,1'-[(Heptane-1,7-diyl)dioxy]bis[(11S,11aS)-10-(tert-butyloxycarbonyl)-7-methoxy-11-(tetrahydro-pyran-2-yloxy)-1,2,3,10,11,11a-hexahydro-5H-pyrrolo[2,1-c][1,4]benzodiazepine-5-one] (**15e**)*

1,7-Dibromoheptane (57.5 mg, 0.22 mmol, 0.5 equiv) was added to the mixture of monomer **14** (0.2 g, 0.44 mmol, 1.0 equiv), potassium carbonate (0.98 mmol, 2.2 equiv) and a catalytic amount of potassium iodide in dry DMF (30 mL), and the resulting mixture was heated to 90°C under a nitrogen atmosphere for 5 h. Removal of excess solvent under reduced pressure afforded a crude solid, which was subjected to flash column chromatography (SiO_2, 50%EtOAc-hexane) to afford the dimerized compound **15e** (200 mg, 0.20 mmol, 91% yield, mixture of diastereomers from THP protecting group) as a solid: $[\alpha]^{20}_D$ = +34° (*c* = 0.13, $CHCl_3$); ^{1}H NMR ($CDCl_3$, 400 MHz): δ 1.25-1.64 (m, 64H, 14-H, 15-H, Boc, THP), 1.67-1.92 (m, 16H, 13-H, THP), 1.93-2.19 (m, 16H, 1-H, 2-H), 3.41-3.73 (m, 16H, 3-H, 11a-H, THP), 3.86-4.08 (m, 24H, 12-H, 7-OMe, THP), 5.01-5.17 (m, 4H, THP), 5.68-5.76 (d, 2H, 11-H), 5.78-5.89 (d, 2H, 11-H), 6.49(s, 2H, 9-H), 6.85 (s, 2H, 9-H), 7.18 (s, 2H, 6-H), 7.22 (s, 2H, 6-H); ^{13}C NMR ($CDCl_3$, 400 MHz): δ 19.8, 20.5, 23.1, 23.3, 25.3, 25.9, 28.1, 28.2, 28.9, 29.1, 29.2, 29.3, 30.9, 31.2, 46.3, 56.1, 56.2, 60.0, 60.2, 63.3, 64.5, 68.9, 69.1, 80.9, 81.3, 88.2, 91.2, 95.7, 100.3, 110.1, 110.6, 114.4, 115.0, 121.0, 129.8, 133.4, 148.4, 148.8, 155.1, 167.4, 167.6; IR (neat): 2942, 1704, 1643, 1605, 1513, 1454, 1432, 1327, 1272, 1202, 1164, 1023cm^{-1}; MS (FAB) *m/z* (relative intensity) 1015 ([*M* + Na]$^{+\cdot}$, 23), 993 (*M* $^{+\cdot}$, 100), 891 (34), 791 (25).

*(b) 1,1'-[(Heptane-1,7-diyl)dioxy]bis[(11aS)-7-methoxy-1,2,3,11a-tetrahydro-5H-pyrrolo[2,1-c][1,4]benzodiazepine-5-one] (**16e**)*

95% TFA (3 mL) was added drop-wise to dimer compound **15e** (200 mg, 0.2 mmol) at 0°C. This was then stirred for 1hr and the mixture was poured into saturated $NaHCO_3$ (30 mL) solution to naturalize the reaction mixture. The mixture was extracted with chloroform (3x20 mL). The organic layer was then washed water (20 mL), brine (20 mL) then dried ($MgSO_4$) and filtrated. The excess solvent was removed under reduced pressure to give the crude product, which was subjected to flash column chromatography (SiO_2, 2%methanol-chloroform). Removal of excess eluent under reduced pressure without heating afforded the final product **16e** (100 mg, 0.17 mmol, 85%) as a solid: $[\alpha]^{20}_D$ = +484° (*c* = 0.15, $CHCl_3$); ^1H NMR ($CDCl_3$, 400 MHz): δ 1.39-1.56 (m, 6H, 14-H, 14'-H, 15-H), 1.83-1.94 (m, 4H, 13-H, 13'-H), 2.00-2.10 (m, 4H, 2-H, 2'-H), 2.26-2.38 (m, 4H, 1-H, 1'-H), 3.54-3.63 (m, 2H, 3-H, 3'-H), 3.70-3.77 (m, 2H, 11a-H, 11a'-H), 3.79-3.91 (m, 2H, 3-H, 3'-H), 3.93 (s, 6H, 7-OMe, 7'-OMe), 4.00-4.18 (m, 4H, 12-H, 12'-H), 6.79 (s, 2H, 9-H, 9'-H), 7.51 (s, 2H, 6-H, 6'-H), 7.65 (d, 2H, *J* = 4, 11-H, 11'-H); ^{13}C NMR ($CDCl_3$, 400 MHz): δ 24.1, 25.8, 28.8, 29.0, 46.6, 53.7, 56.1, 68.9, 110.4, 111.6, 120.0, 140.6, 147.8, 150.9, 162.3, 164.6; IR (neat): 3317, 2936, 1620, 1599, 1505, 1453, 1430, 1381, 1261, 1216, 1092, 1020cm^{-1}; MS (FAB) *m/z* (relative intensity) 653 ($[M + 2 \times MeOH]^{+\cdot}$, 4), 621 ($[M + MeOH]^{+\cdot}$, 3), 589 ($M^{+\cdot}$, 100).

Example 18 (n=8)

*(a) 1,1'-[(Octane-1,8-diyl)dioxy]bis[(11S,11aS)-10-(tert-butyloxycarbonyl)-7-methoxy-11-(tetrahydro-pyran-2-yloxy)-1,2,3,10,11,11a-hexahydro-5H-pyrrolo[2,1-c][1,4]benzodiazepine-5-one] (**15f**)*

1,8-Diiodooctane (81.6 mg, 0.22 mmol, 0.5 equiv) was added to the mixture of monomer **14** (0.2 g, 0.44 mmol, 1.0 equiv) and potassium carbonate (0.98 mmol, 2.2 equiv) in dry DMF (30 mL), and the resulting mixture was heated to 90°C under a nitrogen atmosphere for 5 h. Removal of excess solvent under reduced pressure afforded a crude solid, which was subjected to flash column chromatography

(SiO_2, 50%EtOAc-hexane) to afford the dimerized compound **15f** (190 mg, 0.18 mmol, 85% yield, mixture of diastereomers from THP protecting group) as a solid: $[\alpha]^{20}_D$ = +28° (*c* = 0.16, $CHCl_3$); ^1H NMR ($CDCl_3$, 400 MHz): δ 1.22-1.64 (m, 68H, 14-H, 15-H, Boc, THP), 1.67-1.91 (m, 16H, 13-H, THP), 1.93-2.19 (m, 16H, 1-H, 2-H), 3.41-3.72 (m, 16H, 3-H, 11a-H, THP), 3.85-4.08 (m, 24H, 12-H, 7-OMe, THP), 5.01-5.17 (m, 4H, THP), 5.68-5.76 (d, 2H, 11-H), 5.78-5.89 (d, 2H, 11-H), 6.49(s, 2H, 9-H), 6.85 (s, 2H, 9-H), 7.18 (s, 2H, 6-H), 7.22 (s, 2H, 6-H); ^{13}C NMR ($CDCl_3$, 400 MHz): δ 19.8, 20.5, 23.1, 23.2, 25.3, 25.9, 28.1, 28.2, 28.9, 29.00, 29.04, 29.1, 29.3, 30.9, 31.2, 46.3, 56.1, 56.2, 60.0, 60.2, 63.3, 64.5, 68.9, 69.2, 80.9, 81.3, 88.2, 91.2, 95.7, 100.3, 110.1, 110.6, 114.4, 115.0, 126.4, 129.8, 133.4, 148.5, 148.8, 155.1, 167.4, 167.6; IR (neat): 2940, 1703, 1642, 1604, 1513, 1454, 1432, 1327, 1272, 1202, 1163, 1023cm^{-1}; MS (FAB) *m/z* (relative intensity) 1029 ($[M + Na]^{+\cdot}$, 54), 1007 ($M^{+\cdot}$, 100), 905 (28), 805 (20).

*(b) 1,1'-[(Octane-1,8-diyl)dioxy]bis[(11aS)-7-methoxy-1,2,3,11a-tetrahydro-5H-pyrrolo[2,1-c][1,4]benzodiazepine-5-one] (**16f**)*

95% TFA (3 mL) was added drop-wise to dimer compound **15f** (190 mg, 0.18 mmol) at 0°C. This was then stirred for 1hr and the mixture was poured into saturated $NaHCO_3$ (30 mL) solution to naturalize the reaction mixture. The mixture was extracted with chloroform (3x20 mL). The organic layer was then washed water (20 mL), brine (20 mL) then dried ($MgSO_4$) and filtrated. The excess solvent was removed under reduced pressure to give the crude product, which was subjected to flash column chromatography (SiO_2, 2%methanol-chloroform). Removal of excess eluent under reduced pressure without heating afforded the final product **16f** (105 mg, 0.17 mmol, 96%) as a solid: $[\alpha]^{20}_D$ = +1330° (*c* = 0.13, $CHCl_3$); ^1H NMR ($CDCl_3$, 400 MHz): δ 1.32-1.58 (m, 8H, 14-H, 14'-H, 15-H, 15'-H), 1.78-1.92 (m, 4H, 13-H, 13'-H), 1.93-2.10 (m, 4H, 2-H, 2'-H), 2.26-2.38 (m, 4H, 1-H, 1'-H), 3.55-3.64 (m, 2H, 3-H, 3'-H), 3.70-3.76 (m, 2H, 11a-H, 11a'-H), 3.77-3.89 (m, 2H, 3-H, 3'-H), 3.93 (s, 6H, 7-OMe, 7'-OMe), 3.98-4.15 (m, 4H, 12-H, 12'-H), 6.79 (s, 2H, 9-H, 9'-H), 7.51 (s, 2H, 6-H, 6'-H), 7.65 (d, 2H, *J* = 4.4, 11-H, 11'-H); ^{13}C

NMR ($CDCl_3$, 400 MHz): δ 24.1, 25.8, 28.8, 29.2, 29.6, 46.6, 53.7, 56.1, 69.0, 110.4, 111.6, 120.0, 140.6, 147.8, 150.9, 162.2, 164.67; IR (neat): 3326, 2934, 1599, 1505, 1455, 1431, 1382, 1261, 1216, 1092, 1019cm^{-1}; MS (FAB) *m/z* (relative intensity) 667 ([*M* + 2 x MeOH]$^{+\cdot}$, 3), 635 ([*M* + MeOH]$^{+\cdot}$, 8), 603 (*M* $^{+\cdot}$, 100).

Example 19 (n=9)

*(a) 1,1'-[(Nonane-1,9-diyl)dioxy]bis[(11S,11aS)-10-(tert-butyloxycarbonyl)-7-methoxy-11-(tetrahydro-pyran-2-yloxy)-1,2,3,10,11,11a-hexahydro-5H-pyrrolo[2,1-c][1,4]benzodiazepine-5-one] (**15g**)*

1,9-Dibromononane (63.7 mg, 0.22 mmol, 0.5 equiv) was added to the mixture of monomer **14** (0.2 g, 0.44 mmol, 1.0 equiv) potassium carbonate (0.98 mmol, 2.2 equiv) and a catalytic amount of potassium iodide in dry DMF (30 mL), and the resulting mixture was heated to 90°C under a nitrogen atmosphere for 5 h. Removal of excess solvent under reduced pressure afforded a crude solid, which was subjected to flash column chromatography (SiO_2, 50%EtOAc-hexane) to afford the dimerized compound **15g** (200 mg, 0.19 mmol, 89% yield, mixture of diastereomers from THP protecting group) as a solid: $[\alpha]^{20}_D$ = +43° (*c* = 0.16, $CHCl_3$); ^{1}H NMR ($CDCl_3$, 400 MHz): δ 1.22-1.67 (m, 72H, 14-H, 15-H, 16-H, Boc, THP), 1.68-1.91 (m, 16H, 13-H, THP), 1.92-2.19 (m, 16H, 1-H, 2-H), 3.41-3.74 (m, 16H, 3-H, 11a-H, THP), 3.85-4.09 (m, 24H, 12-H, 7-OMe, THP), 5.01-5.17 (m, 4H, THP), 5.68-5.76 (d, 2H, 11-H), 5.78-5.89 (d, 2H, 11-H), 6.49(s, 2H, 9-H), 6.85 (s, 2H, 9-H), 7.18 (s, 2H, 6-H), 7.22 (s, 2H, 6-H); ^{13}C NMR ($CDCl_3$, 400 MHz): δ 19.8, 20.5, 23.2, 23.3, 25.3, 25.9, 28.1, 28.2, 28.9, 29.01, 29.06, 29.1, 29.4, 30.9, 31.3, 46.3, 56.1, 56.2, 60.0, 60.2, 63.3, 64.5, 69.0, 69.2, 80.9, 81.3, 88.2, 91.2, 95.7, 100.3, 110.1, 110.6, 114.5, 115.0, 126.4, 129.8, 133.4, 148.5, 148.8, 155.1, 167.4, 167.6; IR (neat): 2939, 1703, 1642, 1604, 1513, 1454, 1432, 1327, 1271, 1202, 1163, 1023cm^{-1}; MS (FAB) *m/z* (relative intensity) 1043 ([*M* + Na]$^{+\cdot}$, 45), 1021 (*M* $^{+\cdot}$, 100), 919 (28), 819 (19).

*(b) 1,1′-[(Nonane-1,9-diyl)dioxy]bis[(11aS)-7-methoxy-1,2,3,11a-tetrahydro-5H-pyrrolo[2,1-c][1,4]benzodiazepine-5-one] (**16g**)*

95% TFA (3 mL) was added drop-wise to dimer compound **15g** (200 mg, 0.19 mmol) at 0°C. This was then stirred for 1hr and the mixture was poured into saturated $NaHCO_3$ (30 mL) solution to naturalize the reaction mixture. The mixture was extracted with chloroform (3x20 mL). The organic layer was then washed water (20 mL), brine (20 mL) then dried ($MgSO_4$) and filtrated. The excess solvent was removed under reduced pressure to give the crude product, which was subjected to flash column chromatography (SiO_2, 2%methanol-chloroform). Removal of excess eluent under reduced pressure without heating afforded the final product **16g** (98 mg, 0.15 mmol, 83%) as a solid: $[\alpha]^{20}_D$ = +864° (*c* = 0.14, $CHCl_3$); ^1H NMR ($CDCl_3$, 400 MHz): δ 1.32-1.52 (m, 10H, 14-H, 14′-H, 15-H, 15′-H, 16-H), 1.83-1.90 (m, 4H, 13-H, 13′-H), 2.02-2.09 (m, 4H, 2-H, 2′-H), 2.27-2.36 (m, 4H, 1-H, 1′-H), 3.54-3.64 (m, 2H, 3-H, 3′-H), 3.70-3.77 (m, 2H, 11a-H, 11a′-H), 3.78-3.88 (m, 2H, 3-H, 3′-H), 3.93 (s, 6H, 7-OMe, 7′-OMe), 3.99-4.15 (m, 4H, 12-H, 12′-H), 6.80 (s, 2H, 9-H, 9′-H), 7.51 (s, 2H, 6-H, 6′-H), 7.65 (d, 2H, *J* = 4.4, 11-H, 11′-H); ^{13}C NMR ($CDCl_3$, 400 MHz): δ 24.1, 25.8, 28.8, 29.2, 29.3, 29.6, 46.6, 53.7, 56.1, 69.0, 110.4, 111.6, 120.0, 140.6, 147.8, 150.9, 162.8, 164.6; IR (neat): 2934, 1622, 1599, 1557, 1505, 1455, 1429, 1382, 1339, 1260, 1216, 1092, 1020cm^{-1}; MS (FAB) *m/z* (relative intensity) 681 ([*M* + 2 x MeOH]$^{+\cdot}$, 6), 649 ([*M* + MeOH]$^{+\cdot}$, 12), 617 (*M*$^{+\cdot}$, 100).

Example 20 (n=10)

*(a) 1,1′-[(Decane-1,10-diyl)dioxy]bis[(11S,11aS)-10-(tert-butyloxycarbonyl)-7-methoxy-11-(tetrahydro-pyran-2-yloxy)-1,2,3,10,11,11a-hexahydro-5H-pyrrolo[2,1-c][1,4]benzodiazepine-5-one] (**15h**)*

1,10-Diiododecane (87.8 mg, 0.22 mmol, 0.5 equiv) was added to the mixture of monomer **14** (0.2 g, 0.44 mmol, 1.0 equiv) and potassium carbonate (0.98 mmol, 2.2 equiv) in dry DMF (30 mL), and the resulting mixture was heated to 90°C under a nitrogen atmosphere for 5 h. Removal of excess solvent under reduced pressure afforded

a crude solid, which was subjected to flash column chromatography (SiO_2, 50%EtOAc-hexane) to afford the dimerized compound **15h** (180 mg, 0.17 mmol, 79% yield, mixture of diastereomers from THP protecting group) as a solid: $[\alpha]^{20}_D$ = +59° (*c* = 0.16, $CHCl_3$); ^1H NMR ($CDCl_3$, 400 MHz): δ 1.22-1.67 (m, 76H, 14-H, 15-H, 16-H, Boc, THP), 1.68-1.91 (m, 16H, 13-H, THP), 1.92-2.19 (m, 16H, 1-H, 2-H), 3.41-3.72 (m, 16H, 3-H, 11a-H, THP), 3.85-4.09 (m, 24H, 12-H, 7-OMe, THP), 5.01-5.17 (m, 4H, THP), 5.68-5.76 (d, 2H, 11-H), 5.78-5.89 (d, 2H, 11-H), 6.50 (s, 2H, 9-H), 6.85 (s, 2H, 9-H), 7.18 (s, 2H, 6-H), 7.21 (s, 2H, 6-H); ^{13}C NMR ($CDCl_3$, 400 MHz): δ 19.8, 20.5, 23.2, 23.3, 25.3, 26.0, 28.1, 28.2, 28.9, 29.00, 29.04, 29.1, 29.4, 29.5, 30.9, 31.3, 46.3, 56.1, 56.2, 60.0, 60.2, 63.3, 64.5, 69.0, 69.2, 80.9, 81.3, 88.2, 91.2, 95.7, 100.3, 110.1, 110.6, 114.5, 115.0, 126.4, 129.8, 133.4, 148.5, 148.8, 155.1, 167.4, 167.6; IR (neat): 2938, 1703, 1642, 1605, 1513, 1454, 1432, 1327, 1271, 1202, 1163, 1023cm^{-1}; MS (FAB) *m/z* (relative intensity) 1057 ($[M + Na]^{+\cdot}$, 48), 1035 ($M^{+\cdot}$, 100), 933 (26), 833 (20).

*(b) 1,1'-[(Decane-1,10-diyl)dioxy]bis[(11aS)-7-methoxy-1,2,3,11a-tetrahydro-5H-pyrrolo[2,1-c][1,4]benzodiazepine-5-one] (**16h**)*

95% TFA (3 mL) was added drop-wise to dimer compound **15h** (180 mg, 0.17 mmol) at 0°C. This was then stirred for 1hr and the mixture was poured into saturated $NaHCO_3$ (30 mL) solution to naturalize the reaction mixture. The mixture was extracted with chloroform (3x20 mL). The organic layer was then washed water (20 mL), brine (20 mL) then dried ($MgSO_4$) and filtrated. The excess solvent was removed under reduced pressure to give the crude product, which was subjected to flash column chromatography (SiO_2, 2%methanol-chloroform). Removal of excess eluent under reduced pressure without heating afforded the final product **16h** (103 mg, 0.16 mmol, 96%) as a solid: $[\alpha]^{20}_D$ = +500° (*c* = 0.11, $CHCl_3$); ^1H NMR ($CDCl_3$, 400 MHz): δ 1.26-1.50 (m, 12H, 14-H, 14'-H, 15-H, 15'-H, 16-H, 16'-H), 1.82-1.91 (m, 4H, 13-H, 13'-H), 1.98-2.09 (m, 4H, 2-H, 2'-H), 2.25-2.35 (m, 4H, 1-H, 1'-H), 3.54-3.61 (m, 2H, 3-H, 3'-H), 3.69-3.73 (m, 2H, 11a-H, 11a'-H), 3.78-3.83 (m, 2H, 3-H, 3'-H), 3.93 (s, 6H, 7-OMe, 7'-OMe), 3.98-4.14 (m, 4H, 12-H, 12'-H), 6.79 (s,

2H, 9-H, 9'-H), 7.50 (s, 2H, 6-H, 6'-H), 7.65 (d, 2H, J = 4.4, 11-H, 11'-H); ^{13}C NMR ($CDCl_3$, 400 MHz): δ 24.1, 25.8, 28.8, 29.3, 29.4, 29.6, 46.6, 53.7, 56.1, 69.0. 110.4, 111.6, 120.0, 140.6, 147.8, 150.9, 162.3, 164.6; IR (neat): 2928, 1599, 1557, 1505, 1455, 1430, 1382, 1261, 1216, 1092, 1019cm^{-1}; MS (FAB) *m/z* (relative intensity) 695 ([M + 2 x MeOH]$^{+\cdot}$, 8), 663 ([M + MeOH]$^{+\cdot}$, 16), 631 ($M^{+\cdot}$, 100).

Example 21 (n=11)

*(a) 1,1'-[(Undecane-1,11-diyl)dioxy]bis[(11S,11aS)-10-(tert-butyloxycarbonyl)-7-methoxy-11-(tetrahydro-pyran-2-yloxy)-1,2,3,10,11,11a-hexahydro-5H-pyrrolo[2,1-c][1,4]benzodiazepine-5-one] (**15i**)*

1,11-Dibromoundecane (70.0 mg, 0.22 mmol, 0.5 equiv) was added to the mixture of monomer **14** (0.2 g, 0.44 mmol, 1.0 equiv) potassium carbonate (0.98 mmol, 2.2 equiv) and a catalytic amount of potassium iodide in dry DMF (30 mL), and the resulting mixture was heated to 90°C under a nitrogen atmosphere for 5 h. Removal of excess solvent under reduced pressure afforded a crude solid, which was subjected to flash column chromatography (SiO_2, 50%EtOAc-hexane) to afford the dimerized compound **15i** (210 mg, 0.20 mmol, 91% yield, mixture of diastereomers from THP protecting group) as a solid: $[\alpha]^{20}_D$ = +33° (*c* = 0.18, $CHCl_3$); ^{1}H NMR ($CDCl_3$, 400 MHz): δ 1.22-1.66 (m, 80H, 14-H, 15-H, 16-H, 17-H, Boc, THP), 1.67-1.91 (m, 16H, 13-H, THP), 1.92-2.18 (m, 16H, 1-H, 2-H), 3.41-3.72 (m, 16H, 3-H, 11a-H, THP), 3.85-4.08 (m, 24H, 12-H, 7-OMe, THP), 4.99-5.15 (m, 4H, THP), 5.67-5.75 (d, 2H, 11-H), 5.77-5.88 (d, 2H, 11-H), 6.49 (s, 2H, 9-H), 6.85 (s, 2H, 9-H), 7.18 (s, 2H, 6-H), 7.21 (s, 2H, 6-H); ^{13}C NMR ($CDCl_3$, 400 MHz): δ 19.8, 20.5, 23.2, 23.3, 25.3, 26.0, 28.1, 28.2, 28.9, 29.0, 29.1, 29.4, 29.5, 30.9, 31.3, 46.3, 56.1, 56.2, 60.0, 60.2, 63.3, 64.5, 69.0, 69.2, 80.9, 81.3, 88.2, 91.2, 95.7, 100.3, 110.1, 110.6, 114.5, 115.0, 126.4, 129.8, 133.4, 148.5, 148.8, 155.1, 167.4, 167.7; IR (neat): 2936, 1704, 1642, 1605, 1513, 1454, 1432, 1327, 1272, 1202, 1164, 1023cm^{-1}; MS (FAB) *m/z* (relative intensity) 1071 ([M + Na]$^{+\cdot}$, 76), 1049 ($M^{+\cdot}$, 100), 947 (24), 847 (19).

*(b) 1,1'-[(Undecane-1,11-diyl)dioxy]bis[(11aS)-7-methoxy-1,2,3,11a-tetrahydro-5H-pyrrolo[2,1-c][1,4]benzodiazepine-5-one] (**16i**)*

95% TFA (3 mL) was added drop-wise to dimer compound **15i** (210 mg, 0.20 mmol) at 0°C. This was then stirred for 1hr and the mixture was poured into saturated $NaHCO_3$ (30 mL) solution to naturalize the reaction mixture. The mixture was extracted with chloroform (3x20 mL). The organic layer was then washed water (20 mL), brine (20 mL) then dried ($MgSO_4$) and filtrated. The excess solvent was removed under reduced pressure to give the crude product, which was subjected to flash column chromatography (SiO_2, 2%methanol-chloroform). Removal of excess eluent under reduced pressure without heating afforded the final product **16i** (105 mg, 0.16 mmol, 81%) as a solid: $[\alpha]^{20}_D$ = +623° (*c* = 0.13, $CHCl_3$); ^1H NMR ($CDCl_3$, 400 MHz): δ 1.26-1.52 (m, 14H, 14-H, 14'-H, 15-H, 15'-H, 16-H, 16'-H, 17-H), 1.82-1.92 (m, 4H, 13-H, 13'-H), 1.99-2.12 (m, 4H, 2-H, 2'-H), 2.26-2.38 (m, 4H, 1-H, 1'-H), 3.55-3.64 (m, 2H, 3-H, 3'-H), 3.69-3.77 (m, 2H, 11a-H, 11a'-H), 3.78-3.91 (m, 2H, 3-H, 3'-H), 3.94 (s, 6H, 7-OMe, 7'-OMe), 4.99-4.24 (m, 4H, 12-H, 12'-H), 6.80 (s, 2H, 9-H, 9'-H), 7.51 (s, 2H, 6-H, 6'-H), 7.65 (d, 2H, *J* = 4.4, 11-H, 11'-H); ^{13}C NMR ($CDCl_3$, 400 MHz): δ 24.1, 25.9, 28.9, 29.3, 29.4, 29.6, 46.6, 53.7, 56.1, 69.1, 110.4, 111.5, 120.0, 140.6, 147.8, 150.9, 162.3, 164.6; IR (neat): 3321, 2927, 1599, 1505, 1455, 1431, 1382, 1261, 1216, 1092, 1022cm^{-1}; MS (FAB) *m/z* (relative intensity) 709 ($[M + 2 \times MeOH]^{+\cdot}$, 4), 677 ($[M + MeOH]^{+\cdot}$, 11), 645 ($M^{+\cdot}$, 100).

Example 22 (n=12)

*(a) 1,1'-[(Dodecane-1,12-diyl)dioxy]bis[(11S,11aS)-10-(tert-butyloxycarbonyl)-7-methoxy-11-(tetrahydro-pyran-2-yloxy)-1,2,3,10,11,11a-hexahydro-5H-pyrrolo[2,1-c][1,4]benzodiazepine-5-one] (**15j**)*

1,12-Dibromododecane (73.1 mg, 0.22 mmol, 0.5 equiv) was added to the mixture of monomer **14** (0.2 g, 0.44 mmol, 1.0 equiv) potassium carbonate (0.98 mmol, 2.2 equiv) and a catalytic amount of

potassium iodide in dry DMF (30 mL), and the resulting mixture was heated to 90°C under a nitrogen atmosphere for 5 h. Removal of excess solvent under reduced pressure afforded a crude solid, which was subjected to flash column chromatography (SiO_2, 50%EtOAc-hexane) to afford the dimerized compound **15j** (210 mg, 0.19 mmol, 89% yield, mixture of diastereomers from THP protecting group) as a solid: $[\alpha]^{20}_D$ = +50° (*c* = 0.19, $CHCl_3$); ^{1}H NMR ($CDCl_3$, 400 MHz): δ 1.22-1.66 (m, 84H, 14-H, 15-H, 16-H, 17-H, Boc, THP), 1.67-1.91 (m, 16H, 13-H, THP), 1.92-2.19 (m, 16H, 1-H, 2-H), 3.41-3.72 (m, 16H, 3-H, 11a-H, THP), 3.88-4.08 (m, 24H, 12-H, 7-OMe, THP), 5.00-5.15 (m, 4H, THP), 5.67-5.74 (d, 2H, 11-H), 5.78-5.87 (d, 2H, 11-H), 6.49 (s, 2H, 9-H), 6.85 (s, 2H, 9-H), 7.18 (s, 2H, 6-H), 7.21 (s, 2H, 6-H); ^{13}C NMR ($CDCl_3$, 400 MHz): δ 19.8, 20.5, 23.1, 23.3, 25.3, 26.0, 28.1, 28.2, 28.91, 28.98, 29.0, 29.1, 29.4, 29.5, 29.6, 30.9, 31.3, 46.3, 56.1, 56.2, 60.0, 60.2, 63.3, 64.5, 69.0, 69.2, 80.9, 81.3, 88.2, 91.2, 95.7, 100.3, 110.1, 110.6, 114.5, 115.0, 126.4, 129.8, 133.4, 148.5, 148.8, 155.1, 167.4, 167.7; IR (neat): 2932, 1702, 1644, 1604, 1512, 1454, 1431, 1326, 1271, 1202, 1163, 1023cm^{-1}; MS (FAB) *m/z* (relative intensity) 1085 ([*M* + Na]$^{+\cdot}$, 43), 1063 (*M* $^{+\cdot}$, 100), 961 (31), 861 (17).

*(b) 1,1′-[(Dodecane-1,12-diyl)dioxy]bis[(11aS)-7-methoxy-1,2,3,11a-tetrahydro-5H-pyrrolo[2,1-c][1,4]benzodiazepine-5-one] (**16j**)*

95% TFA (3 mL) was added drop-wise to dimer compound **15j** (210 mg, 0.19 mmol) at 0°C. This was then stirred for 1hr and the mixture was poured into saturated $NaHCO_3$ (30 mL) solution to naturalize the reaction mixture. The mixture was extracted with chloroform (3x20 mL). The organic layer was then washed water (20 mL), brine (20 mL) then dried ($MgSO_4$) and filtrated. The excess solvent was removed under reduced pressure to give the crude product, which was subjected to flash column chromatography (SiO_2, 2%methanol-chloroform). Removal of excess eluent under reduced pressure without heating afforded the final product **16j** (112 mg, 0.17 mmol, 89%) as a solid: $[\alpha]^{20}_D$ = +637° (*c* = 0.13, $CHCl_3$); ^{1}H NMR ($CDCl_3$, 400 MHz): δ 1.24-1.51 (m, 16H, 14-H, 14′-H, 15-H, 15′-H, 16-H, 16′-H,

17-H, 17'-H), 1.78-1.93 (m, 4H, 13-H, 13'-H), 1.98-2.11 (m, 4H, 2-H, 2'-H), 2.25-2.38 (m, 4H, 1-H, 1'-H), 3.54-3.64 (m, 2H, 3-H, 3'-H), 3.68-3.76 (m, 2H, 11a-H, 11a'-H), 3.77-3.89 (m, 2H, 3-H, 3'-H), 3.93 (s, 6H, 7-OMe, 7'-OMe), 3.98-4.14 (m, 4H, 12-H, 12'-H), 6.79 (s, 2H, 9-H, 9'-H), 7.50 (s, 2H, 6-H, 6'-H), 7.64 (d, 2H, J = 4.4, 11-H, 11'-H); ^{13}C NMR (CDCl3, 400 MHz): δ 24.1, 25.9, 28.9, 29.3, 29.5, 29.5, 29.6, 46.6, 53.7, 56.1, 69.1, 110.4, 111.5, 120.0, 140.6, 147.8, 150.9, 162.3, 164.6; IR (neat): 3338, 2926, 1599, 1506, 1456, 1431, 1381, 1261, 1216, 1092, 1023cm^{-1}; MS (FAB) *m/z* (relative intensity) 723 ([M + 2 x MeOH]$^{+\cdot}$, 9), 691 ([M + MeOH]$^{+\cdot}$, 15), 659 ($M^{+\cdot}$, 100).

Example 23 - Determination of DNA Cross-Linking ability and *in vitro* Cytotoxicity

(a) DNA Cross-linking

The extent of DNA cross-linking induced by each PBD dimer was determined using the electrophoretic assay method of Hartley, *et al.* (Hartley, J. A., Berardini, M. D., and Souhami, R. L. (1991) *Anal. Biochem. 193*, 131-134) based on the principle that, following complete denaturation of linear pBR322 DNA (~4,300 bp) to the single-stranded (SS) form, an interstrand cross-link results in renaturation to double-stranded (DS) in a neutral gel.

Closed-circular DNA was linearized with HindIII, then dephosphorylated and finally 5'-singly end-labelled using [$\gamma^{32}P$]-ATP and polynucleotide kinase. Reactions containing 30-40 ng of DNA and the test compound were carried out in aqueous TEOA (25 mM triethanolamine, 1mM EDTA, pH 7.2) buffer at 37°C in a final volume of 50 µl for 2 hours. Reactions were terminated by addition of an equal volume of stop solution (0.6 M NaOAc, 20 mM EDTA, 100 µg/ml tRNA) followed by precipitation with ethanol. Following centrifugation, the supernatant was discarded and the pellet dried by lyophilization. Samples were re-suspended in 10 µl of strand separation buffer (30% DMSO, 1 mM EDTA, 0.04% bromophenol blue and 0.04% xylene cylanol) and denatured by heating to 90°C for 2.5

min, followed by immersion in an ice/water bath. Control non-denatured samples were re-suspended in 10 µl of non-denaturing buffer solution (0.6% sucrose, 0.04% bromophenol blue in aqueous TAE buffer [40 mM Tris, 20 mM acetic acid, 2 mM EDTA, pH 8.1]) and loaded directly onto the gel for comparison.

Electrophoresis was carried out for 14-16 h at 40 V using a 0.8% submerged agarose gel (20 × 25 × 0.5 cm) in TAE buffer. Gels were dried under vacuum for 2 hour at 80°C onto one layer each of Whatman 3MM and DE8I filter papers using a BioRad 583 gel dryer. Autoradiographs were obtained after exposure of Hyperfilm-MP film (Amersham plc, U.K.) to the dried gel for either 4 hour with a screen (or over night, without a screen, to obtain a sharper image). Film bands were quantitated using a BioRad GS-670 imaging laser densitometer. Percentage cross-linking was calculated by measuring the total DNA in each lane (summed density for the double-stranded [DS] and single-stranded [SS] bands) relative to the amount of cross-linked DNA (density of DS band alone). A dose-response curve was derived by plotting drug concentration against the determined percentage level of cross-linked DNA which was then analysed to determine the concentration of test compound that results in 50% cross-linked plasmid DNA (XL_{50}).

(b) In vitro cytoxicity

(i) K562 cells

K562 human chronic myeloid leukaemia cells were maintained in RPM1 1640 medium supplemented with 10% fetal calf serum and 2 mM glutamine at 37°C in a humidified atmosphere containing 5% CO_2 and were incubated with a specified dose of drug for 1 hour at 37°C in the dark. The incubation was terminated by centrifugation (5 min, 300 *g*) and the cells were washed once with drug-free medium. Following the appropriate drug treatment, the cells were transferred to 96-well microtiter plates (10^4 cells per well, 8 wells per sample). Plates were then kept in the dark at 37°C in a humidified atmosphere containing 5% CO_2. The assay is based on the ability of viable cells to reduce a yellow soluble tetrazolium

salt, 3-(4,5-dimethylthiazol-2-yl)-2,5-diphenyl-2*H*-tetrazolium bromide (MTT, Aldrich-Sigma), to an insoluble purple formazan precipitate. Following incubation of the plates for 4 days (to allow control cells to increase in number by approximately 10 fold), 20 µL of MTT solution (5 mg/mL in phosphate-buffered saline) was added to each well and the plates further incubated for 5 hours. The plates were then centrifuged for 5 minutes at 300 g and the bulk of the medium pipetted from the cell pellet leaving 10-20 µL per well. DMSO (200 µL) was added to each well and the samples agitated to ensure complete mixing. The optical density was then read at a wavelength of 550 nm on a Titertek Multiscan ELISA plate reader, and a dose-response curve was constructed. For each curve, an IC_{50} value was read as the dose required to reduce the final optical density to 50% of the control value.

(ii) NCI 60 cell screen

The National Cancer Institute (NCI), Bethesda, Maryland, USA has available an *in vitro* cytotoxicity screen which consists of approximately 60 human tumour cell lines against which compounds are tested at a minimum of five concentrations each differing 10-fold. A 48 hour continuous exposure protocol is used, where cell viability or growth is estimated with an SRB protein assay.

The test compounds were evaluated against approximately 60 human tumour cell lines. The NCI screening procedures were described in detail by Monks and co-workers (Monks, A *et al.*, Journal of the National Cancer Institute, 1991, 83, 757). Briefly, cell suspensions were diluted according to the particular cell type and the expected target cell density (5000-40,000 cells per well based on cell growth characteristics), and added by pipette (100 µL) into 96-well microtitre plates. The cells were allowed a preincubation period of 24 hours at 37°C for stabilisation. Dilutions at twice the intended test concentration were added at time zero in 100 µL aliquots to the wells. The test compounds were evaluated at five 10-fold dilutions (10^{-4}, 10^{-5}, 10^{-6}, 10^{-7} and 10^{-8} µM). The test compounds were incubated for 48 hours in 5% CO_2

atmosphere and 100% humidity. The cells were then assayed using the sulphorhodamine B assay. A plate reader was used to read the optical densities and a microcomputer processed the readings into GI_{50} values (in Moles), which is the dosage required to limit cell growth to 50%.

Results

Compound number	IC_{50} (µM)	GI_{50} (µM)
9a	-	8.9
9b	-	28.1
9c	-	10.2
9d	-	4.0
9f	9.72	-
9h	16.13	-
9i	9.00	-
9j	9.12	-
16d	6.43	-